W. M. KECK LABORATORY OF
ENVIRONMENTAL HEALTH ENGINEERING
CALIFORNIA INSTITUTE OF TECHNOLOGY
PASADENA, CALIFORNIA

FINAL REPORT
ON
NASA RESEARCH GRANT NGR-05-002-036
FOR
INVESTIGATION OF BIOCHEMICAL STABILIZATION OF AQUEOUS SOLUTIONS OF ORGANIC COMPOUNDS BY UNSATURATED FLOW THROUGH POROUS MEDIA

January 1969

Supported by
THE NATIONAL AERONAUTICS AND SPACE ADMINISTRATION

W. M. KECK LABORATORY
OF ENVIRONMENTAL HEALTH ENGINEERING
CALIFORNIA INSTITUTE OF TECHNOLOGY
PASADENA, CALIFORNIA

FINAL REPORT
ON
NASA RESEARCH GRANT NGR-05-002-036
FOR

INVESTIGATION OF BIOCHEMICAL STABILIZATION OF AQUEOUS SOLUTIONS OF ORGANIC COMPOUNDS BY UNSATURATED FLOW THROUGH POROUS MEDIA

January 1969

Supported by
THE NATIONAL AERONAUTICS AND SPACE ADMINISTRATION

TABLE OF CONTENTS

TABLE OF CONTENTS (Continued)

LIST OF TABLES

LIST OF TABLES (Continued)

LIST OF FIGURES

CHAPTER I

INTRODUCTION

1.01 Authorization

This investigation was originally authorized and funded by the Director, Office of Grants and Research Contracts, Office of Space Science and Applications, National Aeronautics and Space Administration, on 28 April 1965 for a period of one year. In the original application and subsequent reapplication for this project grant, it was anticipated that the planned investigation would require about three years. The grant was supplemented and the authorization extended through a second year in a letter from the Acting Director, OGRC, OSSA, NASA on 10 March 1966. The third and final year of the project was authorized, at a considerably reduced level of support, in a letter of 2 June 1967 from the Assistant Administrator for University Affairs, NASA.

This project was part of the research program of NASA's Ames Research Center, and technical liaison was conducted by the Life Sciences Branch of ARC, largely through Mr. E. Gene Lyman and Dr. Philip D. Quattrone.

1.02 Purpose and Objectives

It was the broad intent of this project to study and evaluate many of the parameters associated with the intermittent aerobic filtration of aqueous solutions of organic compounds,

especially the major components of urine, in flow through unsaturated porous media.

In manned space flights of long duration it will probably be necessary to purify and reutilize urine and other wastewaters. Many purification processes involving physical, chemical, and electrolytic phenomena have been investigated by other research groups, and several such mechanisms show promise. Evaporative processes appear to be favored at present, but they must be followed by adsorption on activated carbon to remove the trace organic components that volatilize under the evaporative conditions. In view of the fact that 65 to 70 percent of the solids in human urine are organic, it would appear logical to utilize an aerobic biological process to stabilize and mineralize organic waste waters prior to demineralization by a physical or chemical technique, with the hope that such pretreatment would mitigate the need for post-evaporative carbon adsorption.

Aerobic biological treatment systems, such as the activated sludge process and symbiosis by bacterial-algal cultures, have been investigated by several research groups, especially for combinations of urine, washwater, and feces. No one, however, appeared to have studied the stabilization of full-strength urine and washwater (without feces) by intermittent two-phase filtration through fine porous media. This process offers certain inherent advantages in a zero-gravity environment. Although intermittent filtration through sand or soil is known to be an

effective treatment for mixed municipal wastewaters, its ability to produce a stabilized effluent from urine and other liquid wastes remained to be evaluated. Determination of this ability was the major objective of the project.

1.03 Scope and Limitations

It was envisioned originally that sand filter columns, dosed intermittently with settled municipal sewage initially and later with diluted urine, would "ripen" in a month or so by the development of biological growths capable of degrading the complex organic constituents of urine into simpler organic substances and even into oxidized mineralized end-products such as nitrates, sulfates, phosphates, and bicarbonates. Insofar as possible, the extent and rates of these transformations would be measured under various conditions of operation and for various types and sizes of media. Assuming that these reactions would occur by intermittent percolation under gravity conditions, with or without induced air flow, it was a further intent of the project to operate filters of optimum grain size in a horizontal or sloped position to evaluate the action of capillary and molecular forces, especially under the influence of a forced draft of air. Finally, upon the completion of successful experiments with high degrees of stabilization, the filters would be dismantled and examined biologically to identify and enumerate the organisms responsible for effective biodegradation.

This planned scope of the project proved to be unrealistically optimistic. It was predicated on the basic assumption that

well-ripened sand or soil filters would biodegrade full-strength or partially diluted urine as successfully as they do municipal sewage. Unless and until such stabilization could be achieved, under some feasible system of operation, it was futile to proceed with the hydrodynamic or bacteriological aspects of the investigation. As this report will show, there are certain characteristics of urine that militate against its oxidation and mineralization except in very weak concentrations or in dilute mixtures such as municipal sewage. Consequently, the broad scope of the original proposal was never achieved. The investigation, however, may prove to be worthwhile by contributing to the overall knowledge of this subject and by demonstrating some of the limitations of intermittent percolation of strong organic aqueous solutions through porous media.

1.04 Acknowledgments

In addition to the principal investigator (Professor J. E. McKee) the following employees of the California Institute of Technology participated in the research and preparation of the report. Their contributions to the investigations are gratefully acknowledged:

Project Engineers:

Dr. H. G. Schwartz, Jr. (1 Nov. 1965 to 9 Sept. 1966)

Dr. Maria Puerta (13 Sept. 1966 to 7 Oct. 1966)

Mr. Jack R. Livingston (28 Nov. 1966 to 31 Mar. 1968)

Chemists:

Mr. Jesse Watt (5 July 1965 to 18 Mar. 1966)

Mr. Donald F. Markewich (18 April 1966 to 10 Oct. 1967)

Laboratory Aide:

Mr. Scott Jones (30 Aug. 1966 to 23 Aug. 1968)

Secretaries:

Mrs. Marjorie Connely (1 July 1965 to 26 Sept. 1966)

Mrs. Peggy Freeland (4 Oct. 1965 to 30 Sept. 1967)

Mrs. Wendy Bergman(2 Oct. 1967 to 31 Dec. 1968)

Mr. Albert B. Pincince, then a pre-doctoral graduate student, served as acting project engineer from 15 May 1965 to 1 November 1965, and he continued to be closely associated with the project as he completed his thesis research, which was corollary to this investigation (see Chapter IV and Appendix B). He was supported by a Public Health Service Training Grant and completed his degree requirements by 31 July 1967. The contributions of Dr. Pincince to this project are sincerely appreciated.

Chapters V and VI of this report were prepared largely by Mr. Jack Livingston, whose assistance in this respect is acknowledged with thanks. The other chapters of the report were written by the principal investigator. Because it relates so closely to this report, Dr. Pincince's entire thesis is included herewith as Appendix B.

CHAPTER II

CHARACTERISTICS OF WASTEWATERS IN SPACE VEHICLES

The following information is a summary of data available in several papers and reports as referenced. No attempt was made during this project to make complete chemical analyses of the natural urine used in the experiments. It was contributed by young male project staff members and graduate students, with analyses being made only for gross parameters of organic strength, such as chemical oxygen demand, organic and ammonia nitrogen, total solids, etc.

2.01 Water Quantities

The water balance (total use and total waste) for a 70-kg. man in a restricted environment such as a space vehicle has been calculated, estimated, and reported by several references (e.g. 1, 2, 3, 4, 5, 6, 8, 14, 15, 20). For human water intake and output, the following values are considered to be representative of adult males:

Water Intake (per man)	ml/day
Drinking water	1200
Water in food or used for rehydrating food	1000
Water oxidized by metabolism of food	300
Total	2500

Water Output (per man)	ml/day
Urine	1400
Feces (water content)	100
Respiration and perspiration	1000
Total	2500

For flights of intermediate duration (greater than 15 days but less than 6 months), it is expected that feces will be kept separate from urine, and that it will be packaged, disinfected, and stored (or possibly be jettisoned into space). It is also anticipated that all urine will be reclaimed and all insensible water (respiration and perspiration) will be recovered by dehumidification apparatus. From the foregoing table it is evident that the water oxidized by metabolism of food exceeds that stored with the feces by 200 ml per day. In addition, if fuel cells are used for power generation with the concomitant production of water, the total water available in the cabin will increase daily and may become a problem of ultimate disposal or storage. It is recognized, of course, that space vehicles are not perfectly tight and that some internal atmosphere (including water vapor) escapes; but it still may be necessary to jettison some water to prevent unmanageable accumulations.

In addition to the water taken internally, man needs water for personal cleansing, laundry, and perhaps for washing the cabin. The water requirements for these purposes have been estimated (2, 3, 4, 5, 6, 14, 15) at values ranging from 1800 to 15,000 ml per day. For purposes of this report, the washwater

requirement is taken to be 7000 ml per day per man. This water is suitable for reclamation along with that from urine, respiration, and perspiration. Indeed, by serving as a diluent for urine and by providing carbonaceous compounds to help balance the high nitrogenous content of urine, washwater may prove to be a beneficial component of the water cycle.

The condensate from the dehumidification of water from respiration and perspiration is expected to be almost pure distilled water and consequently would not be mixed with urine or washwater for further treatment. The condensate should be suitable for direct reutilization as drinking water or for food rehydration, although it may have to be passed through activated-carbon columns for the removal of trace compounds that cause undesirable tastes or odors.

Excluding the water in feces and that from dehumidification, therefore, the total amount of water requiring treatment is estimated as follows:

Urine	1400 ml/man/day
Washwater	7000 ml/man/day
Total	8400 ml/man/day

2.02 Water Quality Requirements

How pure must water be for use in drinking or for the preparation of food? In fact, purity may not be the best criterion for optimum water quality. Distilled water generally has a flat and undesirable taste which can be improved by the

addition of dissolved gases and modest quantities of mineral salts. Any water reclamation system should aim, therefore, not for absolute purity but rather for optimum quality as judged by palatability and by criteria of physiological response.

Water quality requirements, however, are generally expressed as standards for maximum allowable (or recommended) concentrations. Drinking water standards have been promulgated for several decades by the U. S. Public Health Service, most recently in 1962 (17). These standards are summarized in Table 2.1. Strictly speaking, they apply only to interstate carriers, national parks and forests, military installations, and other facilities subject to Federal quarantine regulations; but they have been adopted by all (or almost all) states and endorsed by the American Water Works Association. In effect, they have the force of law everywhere in the U.S.A. Moreover, since all U.S. space programs are operated or controlled by Federal agencies, one might conclude that the USPHS Drinking Water Standards apply without question. Yet, there is reason to question them, as noted below.

Drinking water standards have also been promulgated by the World Health Organization, both for international guidance (18) and for use in the more advanced European nations (19). The stricter standards are shown in Table 2.1, for purposes of comparison. In some instances (e.g. coliform bacteria, cyanides, alpha emitters), the WHO standards are more stringent than

Table 2.1

COMPARISON OF DRINKING WATER STANDARDS

	Determination	Maximum Concentrations Given by: USPHS	WHO	SSB(f)
I	Bacterial:			
	Coliform bacteria per 100 ml	1.0	0.05(a)	---
		---	1.0 (b)	---
	Total count per ml	---	---	10.0
II	Physical:			
	Turbidity, silica scale units	5.0	---	10.0
	Color, cobalt scale units	15.0	---	15.0
	Odor, threshold odor number	3.0	---	(d)
	Taste	---	---	(d)
	Foaming	---	---	(e)
III	Chemical: mg/liter			
	Alkyl benzene sulfonate	0.5	---	---
	Ammonia	---	0.5 (a)	---
	Arsenic	0.05(c)	0.2 (a, b)	0.5
	Barium	1.0 (c)	---	2.0
	Cadmium	0.01(c)	0.05(a)	0.05
	Chemical oxygen demand	---	---	100.0
	Carbon chloroform extract	0.2	---	---
	Chloride	250.0	350.0 (a)	450.0
	Chromium (hexavalent)	0.05(c)	0.05(a, b)	0.05
	Copper	1.0	3.0 (a)	3.0
	Cyanide	0.2	0.01(a, b)	---
	Fluoride	1.6 - 3.4 (c)	1.5 (a)	2.0
	Iron	0.3	1.0 (b)	---
	Lead	0.05 (c)	0.1 (a, b)	0.2
	Manganese	0.05	0.1 (a)	---
	Nitrate & nitrite, as NO_3	45.0	50.0 (a)	45.0
	Phenolic compounds	0.001	0.001(a)	---
	Selenium	0.01(c)	0.05(a, b)	0.05
	Silver	0.05(c)	---	0.5
	Sulfate	250.0	250.0 (a)	250.0
	Total solids	500.0	1500.0 (b)	1000.0
	Zinc	5.0	5.0 (a)	---
IV	Radiological: pc/liter			
	Radium-226	3	---	---
	Alpha emitters	---	1.0 (a, b)	---
	Strontium-90	10	---	---
	Beta emitters	1000	10.0 (a, b)	---

a. World Health Organization European Standards, 1961
b. WHO International Standards, 1958
c. Mandatory USPHS Standards, 1962. Others are recommended.
d. None objectionable
e. None persistent more than 15 seconds
f. Ad hoc Panel of Space Science Board, NAS/NRC

those of the USPHS; but more often the USPHS standards have lower permissible limits.

"Some consideration should be given to the rationale under which the (USPHS and WHO) standards were established. In all instances, they are extremely conservative. They are designed to protect children from fluoride and nitrates. They protect aquatic life and goldfish in aquariums with respect to chromates and copper. They meet the threshold limits of taste in the case of copper, iron, zinc, and manganese. In short, they are standards of excellence, but not (always) criteria of human health or limits for the maintenance of a healthful condition of man in space. They should definitely not be applied blindly to the determination of water quality to be met by reclamation systems in space. In many instances, short-term exposure to concentrations considerably in excess of the USPHS or WHO standards would produce no measurable detrimental effect" (8).

Recognizing the constraints that governed the promulgation of USPHS and WHO drinking water standards, the Space Science Board (SSB) of the National Academy of Sciences-National Research Council (NAS/NRC) appointed an ad hoc Panel to establish chemical, physical, and biological standards for reclaimed water intended for human consumption on spacecraft. In its report of September, 1967, the Panel recommended the upper limits shown in Table 2.1. No specific limits are given for iron, manganese, or zinc because undesirable concentrations of these minerals would be manifest in unacceptable taste, color,

or turbidity. The limits for substances that have potentially toxic or adverse physiological effects are set, in some instances, from two to ten times as high as the USPHS drinking water standards; but they are considered (by the Panel) to be well within limits of safety for consumption by healthy adults for periods of three years.

The report of the Panel notes ".....the water used in long-term space missions will be recycled through the human system many times during the course of the flight, providing opportunity for continuing concentration of trace materials. Greater stringency in requirements, particularly with regard to biological quality, is needed to maintain requisite wholesomeness in these circumstances.

"On the other hand, a number of the PHS limits on chemical constituents have been based on considerations of potential accumulated dose during a complete lifetime, or have had reference to complete populations, including infants, aged or infirm persons, and other types of persons with minimal resistance. Presumably, participants in space slights will be robust, healthy adults and the period of ingestion for water of space-flight quality will not exceed a few years. A number of the PHS requirements for chemical quality therefore can be relaxed to some extent without significant deterioration in the wholesomeness of the water for the specific conditions of space flight."

The limits of certain chemical constituents as recommended by the SSB Panel are still quite conservative and might well be relaxed to accommodate a reclamation system that is otherwise optimum or acceptable. The limit of 45 mg/l for nitrate and nitrite, recommended by the PHS and retained by the Panel, is predicated on the possibility of methemogloginemia in infants. It might well be raised for adults when nitrates are a possible product of the reclamation process. There is evidence that humans can drink water containing 1.0 mg/l or more of hexavalent chromium for many years without deleterious effect; hence the Panel's adherence to the strict PHS and WHO standards (0.05 mg/l is difficult to support for space systems, especially when it may be advisable to use high-chromium steel in some parts of the water reclamation system.

The major concern of the SSB Panel lay in the area of microbiological quality of water and the use of coliform organisms as indicators of such quality. The accumulation of organic and inorganic materials in a water-recovery system, as a result of continual recycling, might well create a suitable nutrient medium for the growth of microorganisms that produce toxic metabolites such as endotoxins and exotoxins. For this reason, the Panel found no justification for the establishment of standards based on individual types of organisms. It was considered that the goal should be essential sterility and that total counts of aerobic, facultative, and anaerobic organisms would be the best measure

of this condition. "It was considered essential, moreover, that this criterion of essential sterility be applied to all parts of the recovery system beyond the initial phase-separation step and not simply to the finished product water." (16) By "phase-separation step," the Panel presumably envisions an evaporative process that might be followed by adsorption columns of activated carbon. The criterion of essential sterility could not be applied, quite obviously, to any biological process, such as intermittent sand filtration, that might precede an evaporative system.

2.03 Characteristics of Natural Urine.

The physical properties and chemical composition of human urine have been reported by Allen (13), Altman (9), Cushing (11), Spector (12), and others. The Bioastronautics Data Book (1) presents mean values reported by five sources and the ranges given by Altman, from which Table 2.2 has been prepared to show the major constituents by categories. The figure for total solids (60,000 mg/day) is based on a residue from evaporation at 103°C, which contains salts that include oxides and some water of crystallization; hence the total solids is not the sum of the individual electrolytes and organics. The pH value of urine varies widely, from 4.6 to 8.0, and the density ranges from 1.002 to 1.035 mg/liter.

As Table 2.2 shows, the major electrolytes in urine add up to about 11,700 mg/l, with chloride and sodium predominating. On this basis alone, even without concern about the

Table 2.2

APPROXIMATE AVERAGE CONSTITUENTS OF URINE
(after Bioastronautics Data Book, Ref. 1)

Determination	milligrams per day range		mean	Mean concentration mg per liter
Water	-		1,400,000	-
Total solids	55,000	-70,000	60,000	43,000
Major electrolytes:				
Chloride	4,600	- 9,100	7,300	5,200
Sodium	1,750	- 6,580	4,200	3,000
Potassium	1,120	- 3,920	2,380	1,700
Sulfur	357	- 3,400	1,120	800
Phosphorus (as P)	700	- 1,300	840	600
Calcium	43	- 581	230	164
Bicarbonate	35	- 840	140	100
Magnesium	29	- 307	94	67
Silicon	4.2	- 14.0	9.1	6.5
Trace elements:				
Aluminum	0.049	- 0.112	0.077	0.055
Arsenic	0.0	- 0.091	0.023	0.016
Bromine	0.84	- 7.70	2.1	1.5
Copper	0.0	- 0.049	0.035	0.025
Fluorine	0.3	- 7.0	1.54	1.10
Iodine	0.007	- 0.490	0.28	0.19
Iron	0.02	- 1.10	0.49	0.35
Lead	0.004	- 0.15	0.028	0.02
Manganese	0.007	- 0.098	0.052	0.037
Nickel	0.14	- 0.28	0.15	0.11
Selenium	0.0	- 0.14	0.035	0.025
Tin	0.009	- 0.017	0.013	0.010
Zinc	0.11	- 0.50	0.36	0.26

Table 2.2 (cont'd.)

Determination	milligrams per day range		mean	Mean concentration mg per liter
Organics:				
Urea	14,000	-35,000	30,000	21,400
Other N-compounds:	---	---	5,800	4,150
Amino acids:	1,100	- 2,800	2,000	1,430
Glycine	132	- 671	455	325
Histidine	65	- 499	189	135
Aspartic acid	<10	- 259	119	85
Crystine	10	- 200	119	85
Creatinine	1,000	- 3,219	1,610	1,150
Hippuric acid	70	- 2,500	700	500
Ammonia N	210	- 1,000	700	500
Imadazole derivatives	140	- 300	286	204
Uric acid	56	- 1,000	140	100
Misc. N-compounds	---	---	~ 360	~ 257
Organic acids:	---	---	1,300	930
Carbonic acid	147	- 231	189	135
Formic acid	28	- 140	56	40
Citric acid	128	- 1,400	678	484
Lactic acid	50	- 600	210	150
Oxalic acid	1	- 49	35	25
Oxoglutaric acid	20	- 40	22	16
Pyruvic acid	2	- 100	100	71
Misc. acids	---	---	10	7
Misc. organic compounds	---	---	1,456	1,040
Reducing substances	490	- 1,500	1,000	710
Phenols	200	- 636	400	286
Acetone bodies	2	- 24	14	10
Sugars	1	- 56	42	30
Vitamins, metabolites	---	---	40	29
Hormones	---	---	70	50

organic constituents, urine could not be reutilized for human consumption without some form of demineralization. Biochemical oxidation and stabilization of the organic components to nitrates, bicarbonates, sulfates, etc. will increase the mineral content still more.

The organic content of urine is predominantly urea, averaging about 21,400 mg/l or about 78 percent of all organic matter. Other nitrogen compounds represent 15 percent, making the total nitrogenous component about 93 percent of all organic matter.

In this project, urine and its breakdown products were generally assayed not for the specific constituents in Table 2.2 but rather for collective parameters or major constituents, viz Kjeldahl nitrogen, ammonia nitrogen, nitrates, nitrites, urea, creatinine, total residue on evaporation, chlorides, pH, and chemical oxygen demand (COD). As many as nine young males donated natural urine for the project. These samples were collected daily in accordance with rigid rules (e.g. discarding first sample of the day, no donations following indulgence in alcoholic beverages or during periods of sickness or heavy medication), combined daily, frozen at -60°C, later thawed at 40°C, combined on a weekly or biweekly basis to get a typical average analysis, then refrozen at -60°C until needed. Table 2.3 shows a typical average analysis (approximate) for the natural urine used in these experiments.

Table 2.3

APPROXIMATE AVERAGE ANALYSES OF PROJECT URINE

Determination	Mean Concentration mg/liter
Total solids (residue on evaporation)	47,500
Chlorides	6,250
pH value	5.8
Chemical oxygen demand	4,000
Kjeldahl nitrogen, as N	10,400
Ammonia nitrogen, as N	600
Urea nitrogen, as N	9,000
Creatinine, as N	560

A comparison of the values in Table 2.3 with those in Table 2.2, with proper conversion to similar units, shows that the average project urine was quite similar to that presented in the Bioastronautics Data Book (1), and well within the reported ranges.

2.04 Characteristics of Synthetic Urine

As explained hereinafter, it was advisable at times to use a synthetic urine in order to provide standardized reproducible tests. The synthetic urine as finally used was made by adding the following minerals and organic substances to Pasadena tap water (of Colorado River origin) which already contained many balanced minerals and trace elements. The following weights were diluted with tap water to make one liter of synthetic urine:

Calcium sulfate	388 mg
Magnesium sulfate.....	253 mg
Sodium chloride.....	8,000 mg
Potassium sulfate.....	2,800 mg
Potassium dihydrogen phosphate .	2,500 mg
Ammonium carbonate.....	45,000 mg
Glucose.....	3,300 mg

In earlier formulations, 28,000 mg of urea was used as the nitrogen source, but since the urea rapidly hydrolyzed to ammonium ion and bicarbonate, an equivalent amount of ammonium carbonate was substituted. With allowances for the minerals already in Pasadena tap water, the major electrolytes in this synthetic urine closely approximate those in Table 2.2. The computed COD for the synthetic urine is 1760 mg/l and the computed Kjeldahl nitrogen (equal also to the ammonia nitrogen) is 13,100 mg/l.

2.05 Characteristics of Other Wastewaters in Spacecraft

The wastewaters from personal cleansing (sponge baths), laundry, and perhaps some cabin cleaning are estimated at 7000 ml/man/day. The exact nature of wastewater from cabin cleaning was not determined from the literature; but it is assumed that such wastes will not exert a significant carbonaceous or nitrogenous oxygen demand, except for their detergent content as noted below.

Body and laundry wastes will contain body secretions and probably a large amount of detergent. The general magnitude of body secretions (sweat, sebum, and other skin secretions) is shown in Table 2.4. When diluted in 7.0 liters of water, these daily amounts provide concentrations very much lower than those in urine. From Table 2.4, the COD of such wastewaters (without detergents) is estimated to be about 230 mg/l and the Kjeldahl nitrogen is estimated at only about 150 mg/l. However, based on data presented by Wallman et al (21) and by Ingram (22), the active detergent concentration of washwater is estimated to be about 700 mg/l. Detergents typically contain 60-70 percent carbon. Assuming 60 percent carbon, the COD from these added detergents alone is calculated to be 1120 mg/l, and the total COD of the wastewaters will be increased to about 1350 mg/l.

The blending of washwaters with urine would be expected to provide two advantages: (a) dilution, and its consequent lowering of the high urine concentrations, and (b) carbonaceous compounds, largely from detergents, which might serve to improve biochemical stabilization by providing a better carbon-nitrogen ratio. In this project, however, no attempt was made to simulate or synthesize washwaters for mixing with urine. Instead, as noted hereinafter, various dilutions of urine in distilled water were used.

Table 2.4

PROBABLE AMOUNTS OF MATERIAL REMOVED DAILY FROM THE SKIN OF AN AVERAGE ADULT BY WASHING AND BY ABSORPTION IN CLOTHING

(After References 1 and 3)

Component		Amount in mg/day	Concentration in mg/l*
Minerals:-	Total....	2471	353
Sodium	840		
Chloride.....	1250		
Potassium.....	370		
Calcium	10		
Magnesium	1		
Carbonaceous compounds:.....		886	127
Fatty acids.....	420		
Lactic acid.....	250		
Cholesterol.....	75		
Glucose.....	50		
Triglycerides.....	50		
Others	41		
Nitrogenous compounds:		411	59
Urea.....	350		
Amino acids.....	34		
Creatinine.....	20		
Others.....	7		

* When diluted in 7.0 liters of water

CHAPTER III

THE RATIONALE FOR BIOCHEMICAL STABILIZATION OF URINE BY PERCOLATION THROUGH SOIL

Before considering the experimental work of this project, it is well to review and rationalize the state of knowledge dealing with the biodegradation of aqueous solutions of organic compounds in aerobic or anaerobic percolation through soil. The final stabilized products from the aerobic decomposition of organic matter in aqueous solution are carbonates, bicarbonates, nitrates, phosphates, sulfates, other mineral electrolytes, water, and a stable residue of refractory organic substances. Anaerobic degradation yields carbon dioxide or bicarbonates, ammonia or amino acids, methane, sulfides, other reduced substances, and a humus-like putrescible residue. The aim of this project was to achieve a high degree of aerobic decomposition.

3.01 Hydrolysis of Urea and Other Nitrogenous Compounds

Urea, by far the most abundant organic constituent of urine, is hydrolyzed rapidly and completely at 20-25°C to carbon dioxide and ammonia, or more specifically to ammonium and bicarbonate ions (or carbonate ions, depending on the resultant pH). This hydrolysis is catalyzed by the enzyme "urease," which is produced by the ubiquitous bacterium *Micrococcus ureae* and other organisms (27, 28). Thus:

$$NH_2CONH_2 + H_2O \longrightarrow CO_2 + 2NH_3 \quad \ldots\ldots\ldots (3.1)$$

or more specifically, since the CO_2 and NH_3 both react with water:

$$NH_2CONH_2 + 3H_2O \longrightarrow HCO_3^- + 2NH_4^+ + OH^- \quad \ldots\ldots (3.2)$$

This reaction does not require atmospheric or dissolved oxygen. It occurs rapidly at room temperature as soon as urine is seeded by Micrococcus ureae or other ammonifying organisms. Furthermore, the hydrolysis raises the pH value by producing excess hydroxyl ions and it provides a large buffering capacity related to the dissociation constants for bicarbonates and ammonium hydroxide. In this project, the pH value of the natural urine has generally varied from 5.5 to 6.3 whereas the hydrolyzed percolate from soil columns in which nitrification did not occur had pH values of 8.5 to 9.3, with most values about 8.8. Since urea, as such, is seldom present in the percolate and the pH has risen, it is evident that conversion of urea to ammonium and bicarbonate (or carbonate) ions is essentially completed in the short period of percolation in the columns.

Whether the nitrogen in the hydrolyzed product is present as ammonium ion, as ammonium hydroxide, or escapes as ammonia gas depends on the pH value and the conditions for gas exchange. Thus:

$$NH_4^+ + OH^- \rightleftharpoons NH_4OH \rightleftharpoons \underset{(gas)}{NH_3\uparrow} + H_2O \quad \ldots\ldots (3.3)$$

The dissociation constant for ammonium hydroxide is 1.8×10^{-5} at $25^{\circ}C$. At pH 7, the ratio of ammonium ions to undissociated ammonium hydroxide is 180 to 1 and hence almost all nitrogen exists in the form of ammonium ions. But at pH 9, which results from hydrolysis of urea in urine, the ratio is only 1.8

to 1; hence about 35 percent of the nitrogen is in the form of undissociated ammonium hydroxide from which, under conditions optimum for gas transfer, gaseous ammonia may be released. This relationship accounts for the strong smell of ammonia at cattle feed pens and other places where urine is discharged to the soil.

The second most abundant nitrogenous compound in urine, creatinine, is also hydrolyzed rapidly to ammonium ions in the presence of dissolved oxygen, thus:

$$CH_3NC(:NH)NHCOCH_2 + 5H_2O + 3O_2 \longrightarrow$$

$$\longrightarrow 4HCO_3^- + 3NH_4^+ + H^+ \quad \ldots\ldots\ldots\ldots\ldots\ldots (3.4)$$

By producing hydrogen ions, this reaction tends to decrease the pH value; but any such change is more than offset by the overwhelming effect of the hydrolysis of urea. In this project, creatinine was seldom found in the percolate, so it was apparently completely hydrolyzed in a short time.

Similarly, glycine and other amino acids are hydrolyzed to ammonium ions and to acetic or higher organic acids which may be further oxidized to bicarbonates and water. The release of ammonia from any proteinaceous compound is known as "ammonification." It is carried out by a number of different species of microorganisms, among which Bacillus mycoides and Proteus vulgaris are prominent. It is apparent, therefore, that the simple predominant nitrogeneous compounds in urine are rapidly hydrolyzed with the production of ammonium ion or

undissociated ammonium hydroxide.

3.02 Nitrification of Ammonia

The metabolism of inorganic nitrogen is described by McElroy and Glass (23), Quastel and Scholefield (24) and by many others. Only the highlights applicable to this project are summarized in this section.

The oxidation of ammonia proceeds in two stages. One group of organisms belonging to the genera Nitrosomonas and Nitrosococcus oxidize ammonium ion to nitrites, thus:

$$2NH_4^+ + 3O_2 \longrightarrow 2NO_2^- + 2H_2O + 4H^+ \quad \ldots\ldots\ldots (3.5)$$

These bacteria are autotrophic, obtaining their energy from the oxidation above and synthesizing their protoplasm entirely from inorganic salts, especially ammonium and bicarbonate ions. They are strict aerobes and die off rapidly in the absence of oxygen. A second group of bacteria, belonging to the genus Nitrobacter, oxidize nitrites to nitrates, thus:

$$2NO_2^- + O_2 \longrightarrow 2NO_3^- \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.6)$$

These organisms are also autotrophic and strict aerobes. The optimum temperature for nitrification in temperate regions is approximately 25°C.

From equation 3.5 above, it is noted that the production of each molecule of nitrite is accompanied by the release of two hydrogen ions. It is obvious, therefore, that the pH value will fall progressively during nitrification unless there is good buffering. Indeed, when one molecule of urea is hydrolyzed to

$2NH_4^+$ (equation 3.2) one hydroxyl ion is released; but when the same amount of nitrogen is subsequently oxidized to nitrite, four hydrogen ions are produced, making a net gain of three hydrogen ions. Subsequent oxidation to nitrate, however, does not further alter the pH value. It is to be expected, therefore, that nitrification will be accompanied by a lowering of the pH value, the extent of which will depend on the buffering capacity of the system, including the calcium carbonate content of the soil. Indeed, a drop in the pH value was found in this project to be a good indication that nitrification was occurring.

Unfortunately the key genus in this transformation, Nitrosomonas, is very sensitive to an optimum pH value of about 8.2-8.7. Its rate of oxidation is halved when the pH drops to 7.0 or rises to 9.3 according to Hofman and Lees (32), and it is almost ineffective below pH 6.0 or above pH 9.5, according to Meyerhof (33). In a poorly buffered system, therefore, this organism is self-inhibiting. Nitrobacter is somewhat more tolerant, functioning well between pH values of 7 to 10 and marginally down to pH 6. It will have no source of nitrites, however, if the Nitrosomonas bacteria cannot operate.

The rate and extent of nitrification are greatly enhanced by the presence of large surface areas or interfaces. Indeed, nitrification in the laboratory and the isolation of the responsible organisms was first accomplished in 1877 by means of a perfusion technique comparable to intermittent sand filtration, i.e.

sewage was applied once a day to a column of sterile sand and chalk. It has since been demonstrated that soil is a far better medium for supporting nitrification than is sand (24). It is to be expected, therefore, that nitrification will be favored by the highest ratio of surface area to unit volume, provided that the system can be kept aerobic.

The rate of nitrification is a function of the degree to which ammonium ions are absorbed on, or combined in, the soil in the form of the soil's base-exchange complexes. Nitrifying bacteria grow on the surfaces of soil particles at the sites where ammonium ions are held in base-exchange combinations, and they proliferate at the expense of such adsorbed ammonium ions. The finer the soil particles, the greater is the adsorption and hence the better the nitrification, in an aerobic system. In a given soil, however, a condition is reached eventually when the sites of proliferation are all saturated with nitrifying organisms. This condition determines the maximum capacity of the given soil column for nitrification.

3.03 Inhibitors to Nitrification

In addition to pH values of 7.0 or lower, nitrification of ammonia is also inhibited by certain organic compounds. According to Quastel and Scholefield (24), the following concentrations inhibit the conversion of ammonia to nitrites:

Substance	Inhibiting Concentration, mg/l	Range of Concentration in Urine, mg/l
Thiourea	25	---
Guanadine	214	7- 14
Ethyl urethane	324	---
p-Aminosalicylic acid	460	---
Glycine	980	94- 480
Creatine	1311	0- 570
Creatinine	2260	715-2300

Although the concentrations of thiourea, ethyl urethane, and p-aminosalicylic acid in urine are not reported (1, 9), it is unlikely that they would exceed the inhibiting concentrations shown above. None of the other organic compounds appears to be inhibitory in the range of concentrations reported for urine, except for the extreme upper range for creatinine. Results from this project indicated that creatinine was rapidly hydrolyzed in soil percolation and thus would not be inhibitory.

None of the common inorganic ions appears to be markedly inhibitory to nitrification. At a concentration of 0.1 N, sodium chloride produced about six percent inhibition (corresponding to 2,300 mg/l of sodium and 3,550 mg/l of chloride, or about 70 percent of these salts in average urine). It may be assumed, therefore, that the osmotic pressure of the salts in urine exerts a mild inhibiting action to nitrifying bacteria previously acclimated to fresh water; but this effect should diminish

quickly. Heavy metals are inhibitory at concentrations of 0.005 M, but such concentrations do not occur in urine.

It was believed formerly that organic matter, per se, and especially carbonaceous matter, was deleterious to the growth of nitrifying organisms. Jensen (30), however, showed that a species of Nitrosomonas can oxidize ammonia in the presence of 10 percent glucose or 20 percent sucrose in well-shaken flasks. According to Quastel and Scholefield (24), carbohydrates such as glycerol and glucose cause an initial depressing effect on the rate of nitrification by stimulating the growth of heterotrophic organisms that utilize ammonia, along with the carbon from glucose or glycerol, in cell synthesis. This proliferation of heterotrophs immobilizes the ammonia until the rate of die-off and release of ammonia equals the rate of assimilation. Then, nitrification may be expected to occur. Heavy growths of heterotrophs may also clog the interstices of the soil, especially near the top surface, and thereby impede the penetration of atmospheric oxygen, so necessary for nitrification. Furthermore, the heterotrophs may coat many of the surfaces of soil particles and thereby diminish the rate of adsorption of ammonia. Hence, although carbonaceous organic matter may not be toxic, it may inhibit or delay nitrification. In hydrolyzed urine, however, the amount of available ammonia far outweighs the carbonaceous material so that the temporary immobilization of some ammonia by heterotrophs should not decrease signficantly the

supply of ammonia for Nitrosomonas. It is believed that the clogging and coating phenomena may be the major deleterious effects of carbonaceous matter.

Perhaps the greatest inhibitor to the nitrification of ammonia is a high concentration of ammonia itself. Undissociated ammonium hydroxide at concentrations as low as 1.0 mg/l has been reported as lethal to sensitive aquatic animals. In perfusion columns, however, nitrifying organisms appear to be most active in the presence of ammonium ion at concentrations of 70 mg/l as N. According to Quastel and Scholefield (24), urea, arginine, creatine, and glycine were not inhibitory to nitrification at 0.00364 molar concentrations (corresponding to 51 mg N/liter). According to Hofman and Lees (32), ammonium sulfate at 2.5×10^{-3}M (70 mg/l as N) was oxidized by Nitrosomonas in liquid culture completely in four hours and to about 80 percent completion in two hours. Concentrations of 20 to 30 mg/l of ammonia nitrogen in municipal sewage are readily oxidized to nitrates in extended-aeration activated-sludge plants and also in intermittent sand filters.

Meyerhof (33) reported that the rate of oxidation of ammonia by Nitrosomas was greatest at 70 mg N/liter, diminished to about 40 percent of the optimum rate at 700 mg N/liter, and to about 10 percent at 1400 mg N/liter. Furthermore, the inhibiting effect was much greater at pH 9.4-9.5 than at pH 7.6-7.8, which implies that the toxic agent is the undissociated ammonium hydroxide (or so-called free ammonia).

The highest concentration of urea permitting nitrification, as reported by Jensen (30) was 3.5 percent or 0.58 M, corresponding to 16,300 mg N/liter. This value is considerably higher than the concentration of urea in urine (Table 2.2). On that basis, it would appear that urea and its hydrolysis product, ammonium ion, would not inhibit nitrification of urine during intermittent percolation through soil. Yet, as this project demonstrates, full-strength urine is not readily nitrified.

It has been postulated that the oxidation of ammonia to nitrite proceeds in three steps, with hydroxylamine (H_2NOH) and hyponitrous acid (HNO) as intermediate products. Jensen (30) reported that free hydroxylamine at concentrations as low as 0.0003 M (4.2 mg N/liter) suppressed the nitrification of ammonium sulfate, while Lees (31) claimed that hydroxylamine is not nitrified in soil at concentrations as low as 7.0 mg N/liter. However, at 1.5 mg N/liter, it is nitrified as rapidly as ammonia and thus it is a possible intermediate in the nitrification process. If indeed the nitrification of ammonia proceeds through hydroxylamine and if indeed this intermediate is inhibitory except at very low concentrations, it would appear that nitrification would be very slow and well-nigh impossible. Yet, it takes place readily in percolating sewage.

Inasmuch as the literature on the inhibiting effect of ammonia itself is contradictory, enlightenment on this matter became a major goal of this project.

3.04 Movement of Nitrogenous Compounds in Soil

The literature on this subject has been summarized by Bailey (26), Stout et al (29), and Preul and Schroepfer (25). They show that the ammonium ion (NH_4^+) is readily adsorbed to the negative surfaces of soil particles. Ammonium ions are held to solid-phase surfaces more strongly than sodium ions, but less tightly than calcium or magnesium ions. Under the pH conditions normally found in soils (pH 4-9), ammonium ion is probably adsorbed chemically by clays in an exchangeable form, and by the organic matter in the alkaline range. Apparently, some ions can decrease the adsorption of ammonium ion under certain conditions.

Urea is only weakly ionized and consequently is not highly adsorbed; but since it hydrolyzes rapidly to ammonium ions it does not appear in the leachate (except in sterile systems).

Nitrite and nitrate ion, however, are not adsorbed and are readily leached from soil by percolating water. When nitrifying organisms convert adsorbed ammonium ions to nitrites, the product is readily leached, thereby making more base-exchange sites available for further adsorption of ammonium ions.

It can be expected therefore that ammonium ions added to a column (or urea that is rapidly hydrolyzed to ammonium ions) will be adsorbed up to the exchange capacity of the soil system and then appear in the leachate if nitrification does not occur. Any nitrites and nitrates formed in the column may be expected to appear rapidly in the leachate.

3.05 Removal of Carbonaceous Matter

The carbonaceous matter, as measured by the COD, is largely removed during percolation through soil by adsorptive mechanisms. It is subsequently metabolized under aerobic conditions by heterotrophic organisms to bicarbonates, water, sulfates, etc. This phenomenon has been demonstrated frequently for municipal sewage applied intermittently to sand beds and, as shown by this project, it holds also for urine. Not all of the COD is removed, however, for some of the components are not readily adsorbed and some are not rapidly oxidized.

3.06 Removal of Phosphates

As shown by Table 2.2, phosphorus is a major mineral component of urine. It is also a constituent of most commercial detergents and thus would appear in any washwater that might be mixed with urine prior to reclamation.

In contrast with the rapid leaching of nitrites and nitrates in soil columns, most phosphorus-bearing compounds react vigorously with the soil, such that very little phosphorus appears in the percolate (26). The fixation of phosphorus is governed by the type of soil, particle size, pH, reduction potential, temperature, organic content, and reaction time (detention time) in the column. Fixation is optimum at pH values above 8.0, mainly as calcium phosphate, or at pH values below 5.0, by aluminum, iron, or manganese. The poorest fixation occurs in the pH range of 6.0 to 7.0.

The capacity of a short column to fix and retain phosphorus compounds is obviously finite, and consequently phosphorus can be expected to appear eventually in the percolate. It should occur, however, as a mineralized phosphate inasmuch as the organic phosphorus compounds should be biodegraded during a long retention in the adsorbed state.

No attempts were made to study the removal of phosphorus during this project inasmuch as such removal was of minor interest compared with the major problem of nitrification.

CHAPTER IV

A BRIEF HISTORY OF THE PROJECT

To provide a better understanding of the development and results of this research, a brief chronological resume' is presented in this chapter. It should serve as a prelude to the detailed material of subsequent chapters. It may also enable the hurried reader to skip the detailed account and turn to the summary and conclusions.

4.01 Initial Preparations

The early days of the project, starting in mid-May, 1965 were utilized primarily for the design, construction, and mechanical operation of the laboratory soil columns. The proper construction of these columns required considerable trial and error in order to accomplish intermittent scheduled and measured dosing of liquid, with a simultaneous passage of air or oxygen through the columns. It became necessary to modify the design several times during the three years of the project to correct unanticipated difficulties. The columns are described in detail and illustrated in Chapter V.

Twenty columns of one-inch inside diameter were fabricated initially. Ten columns were packed with 61 cm of silica sand having a geometric mean size (M_g) of 0.56 mm and a geometric standard deviation (S_g) of 1.2. The other ten columns held 61 cm of a finer silica sand, with M_g of 0.12 mm and S_g of 1.14. It became necessary later to abandon the finer sand because it

clogged so rapidly and to repack these columns with the 0.56 mm sand. Also, near the end of the project, sand with M_g = 1.4 mm was employed.

Concurrently with the fabrication of the columns, literature investigations were conducted and preliminary experimentation was undertaken with respect to analyses of urine and its constituents. From these tests and from calculations based on Table 2.2, it became apparent that complete aerobic oxidation of urine would require so much oxygen that normal diffusion of air from the ambient atmosphere into the soil would be inadequate. Provisions were made, therefore, to apply air under pressure and to utilize pure oxygen, if advisable.

A doctoral candidate, Albert B. Pincince, selected for his thesis research one important aspect of this project, viz the diffusion of air into intermittent sand filters. Although he worked with municipal sewage and synthetic percolates rather than urine, his findings are so relevant to this project that they are discussed at several points in this report and his entire thesis is included herewith as Appendix B.

4.02 Ripening of Columns

In order to develp a rich biological flora in the sand columns and to assure an abundance of nitrifying organisms, the columns were dosed initially with settled municipal sewage that had been passed through glass wool to remove coarse particles. From the start, each column functioned effectively to remove

80 percent or more of the COD, largely by adsorption. Over a month was required, however, for the bacterial population to be able to begin to form nitrates from the ammonia and organic nitrogen, and two months or longer transpired before nitrification was developed sufficiently to oxidize about two-thirds of the nitrogen to nitrates.

In November 1965, the column that had been operating longest was connected to a pure oxygen supply. Prior to that time, the top of the column had been open to the atmosphere. It was hoped that an increased oxygen tension would be conducive to further nitrification. The opposite effect was observed, however, for nitrification decreased markedly during the two months that the column received pure oxygen. In January 1966 the pure-oxygen supply was removed and the column was connected to a compressed-air line, following which the ability to nitrify improved.

Ponding occurred in all of the fine-sand filters shortly after dosing began. Infiltration rates were so low that these systems had to be eliminated from practical consideration. Moreover, significant nitrification did not occur until after about five months of operation. These filters were discontinued and the columns repacked with the medium-size sand. No detailed attempt was made, as originally planned, to determine an optimum grain size.

4.03 Application of Natural Urine

Modifications of the columns to improve air flow, repacking some columns with medium-size sand, and further ripening with municipal sewage continued until July 1966 when ten of the columns were placed on a feed of natural urine as follows:

Columns 1 & 2	full-strength urine
Columns 3 & 4	50% urine, 50% distilled water
Columns 5 & 6	20% urine, 80% distilled water
Columns 8 & 9	10% urine, 90% distilled water
Columns 14 & 15	5% urine, 95% distilled water

Other columns continued to be ripened with municipal sewage for future standby.

Each column received 100-150 ml per day of the stated dilution, corresponding to a hydraulic load rate of 20-30 cm per day. About 10 ml of feed water was dosed in a period of two seconds once every two hours, or 12 times each day. This small quantity quickly infiltrated into the sand and the column was subjected almost continuously to a small flux of compressed air. All percolate and all air passing through each column were collected and measured.

It had been anticipated that the sewage-ripened columns would be capable of converting most of the organic nitrogenous compounds in urine to stabilized nitrates, after a reasonable period of acclimatization to the urine feed. Such was not the case, however. The columns at all dilutions were almost 100 percent effective in destroying both urea and creatinine by

hydrolysis or ammonification; but the percolates contained ammonia rather than nitrates, even at the 10:1 dilution.

The Kjeldahl nitrogen for the undiluted urine used at this stage of the project averaged about 10,000 mg N/liter. Based on the inhibitory effects of ammonia reported by Meyerhof (33), as described in Section 3.03, the columns with 20:1 dilution (500 mg N/l) should have shown some nitrification at pH 7.5-8.0; however, the pH values of these percolates averaged about 8.8. It is apparent, therefore, that a toxic action was being exerted by undissociated ammonium hydroxide.

Approximately 4000 ml of air per day was passed through each column. Stoichiometrically, this volume of air should have sufficed for complete nitrification in the 5-percent and 10-percent urine columns. Hence, the overall availability of oxygen does not appear to have been the limiting factor.

In localized parts of the soil columns, however, anaerobiosis may have occurred. During this phase of the project, inorganic precipitates and biological growths slowly clogged some of the columns. The rate and extent of clogging were proportional to the urine concentration in the feed. The clogging action was generally confined to the upper three inches of sand. It was relieved periodically by scarifying the sand with glass tubing.

4.04 Dosing With Synthetic Urine

In view of the clogging problem and difficulties in obtaining nitrogen balances with natural urine, it was decided to experiment with a synthetic urine of known and adjustable composition. The formulation described in Section 2.04 was utilized except that 28,000 mg/l of urea was employed initially as the nitrogen source. After a period of reripening with municipal sewage, synthetic urine was applied at the same dilutions used previously for natural urine. Unfortunately, the results were the same, viz no nitrification, but excessive clogging, both chemical and biological.

Part of the clogging was attributed to a white precipitate that tended to cement the sand particles. Preliminary tests indicated that this precipitate was a calcium phosphate; hence the potassium dehydrogen phosphate was eliminated from the formulation. Clogging persisted, but it appeared to be mostly biological and was most severe at the highest concentrations of synthetic urine. It was decided, therefore, to confine further tests to the 20:1 and 10:1 dilutions in an attempt to get nitrification at these concentrations of ammonia, either with natural or synthetic urine.

4.05 Oxygen Relationships in Intermittent Sand Filtration

Concurrently with the operation of the laboratory sand columns with natural and synthetic urine, a doctoral candidate was studying oxygen balances and transfer mechanisms for intermittent sand filters. He found that the oxygen content of the

system at sufficiently long times after addition of wastes can be described by a quasi-steady-state diffusion equation, including a term for an oxygen sink.

Shortly after each intermittent liquid dose infiltrates into the sand, the high oxygen tension in the unsaturated pores is governed largely by the displacement phenomenon, or piston action. The oxygen content at each level of the filter then decreases until the rate of diffusion equals the rate of oxygen demand; but since the rate of demand decreases with time, the oxygen tension at a given depth slowly but gradually increases. The major factor controlling this diffusion, however, is the biological clogging, especially in the upper few centimeters of the filter.

4.06 Cattle Feed Pens

It was reasoned that the sandy soil beneath feed pens that had been subjected to urine discharges from scores of cattle per acre for a decade or longer would be well-seeded with nitrifying organisms. Furthermore, with intermittent discharges and the frequent removal of manure, and with its natural porous nature; such soil should be subject to good aeration. A suitable feed pen was selected in the southern part of Los Angeles and core samples were taken to depths of three and four feet. The soil moisture was removed by rinsing and dilution, then analyzed for nitrates. As with the laboratory sand columns, nitrification was not apparent in this natural condition.

4.07 Further Column Tests

Continuing the attempt to get nitrification in the laboratory, four columns were repacked with silica sand with M_g = 1.4 mm instead of M_g = 0.56 mm. Furthermore, all columns of either sand size were dosed with 10:1 dilutions of natural or synthetic urine. In some cases, hydraulic loads were decreased to 80 ml per day or even lower in order to provide a longer retention time or contact period for the soil moisture. In one column, the same percolate was recycled over and over again for about two weeks with the hope that it would eventually nitrify. Carbon dioxide was added to the air stream of one column. Also the concentration of carbonaceous material was varied in the feed to some columns. Natural and synthetic urines were blended with municipal sewage (10 percent) to assure adequate sources of carbon compounds and trace elements. For all of these modifications, very little nitrification was achieved; in fact the highest conversion efficiency of any phase of the experimentation was about 14 percent.

4.08 Use of Other Porous Media

In the last stages of the experimental work (when time, budget, and technician assistance were very limited), two of the columns was repacked with calcium carbonate particles and two with granular activated carbon. Calcium carbonate was selected for its buffering capacity and activated carbon for its powerful adsorptive power.

At a 10:1 dilution of natural urine and a hydraulic loading rate of 200 ml per day, both activated carbon columns removed an average of about two thirds of the Kjeldahl nitrogen applied, with effluent pH values of 3.4-4.2 for one column and 3.7-5.4 for the other. It is evident, therefore, that the tremendous surfaces areas and adsorptive capacity of activated carbon favored nitrification. Further pursuit of this avenue was denied by termination of the project; however, it would be illogical to use granular activated carbon columns for biochemical oxidation of urine prior to a demineralization process. It is much more rational to demineralize by evaporation and then use the activated carbon for taste and odor control. Results with calcium carbonate packing were somewhat erratic and do not lead to definitive conclusions.

4.09 Summary

As this brief history relates, the major objective of the project, i.e. to oxidize nitrogenous and other organic constituents of urine to mineral compounds, was not realized, even at dilutions of 10:1 and 20:1. In all probability, the major inhibitor to nitrification was the high concentration of toxic undissociated ammonium hydroxide at pH values of 8.5-9.2, following hydrolysis of urea to ammonia. Another unfavorable factor was the excessive clogging of the sand near the top of the columns with concomitant detriment to the maintenance of aerobic environments in all parts of the soil moisture.

Under optimum conditions, e.g. proper grain size and characteristics, maintenance of a favorable pH range, adequate aeration, probable recycling of percolate, ideal C:N:P ratios, etc., it might be possible to get complete nitrification in an intermittent sand filter; but the results of this research do not lend much encouragement to this prospect.

CHAPTER V

EXPERIMENTAL APPARATUS AND ANALYTICAL PROCEDURES

This chapter describes the laboratory columns of porous media used for these experiments and some of the problems encountered therewith, along with the palliatives employed. It also presents a brief synopsis of the analytical procedures, including the effects of interfering substances.

5.01 Initial Sand Columns

The laboratory columns were designed so that the dosing schedule could be regulated, so that the air flow could be measured, and so that carbon dioxide production could be determined. A schematic diagram of one intermittent sand filtration apparatus is shown in Figure 5.1 and a photograph of the first 20 columns and appurtenances is presented as Fig. 5.2.

The columns were constructed of clear lucite tubing of one-inch inside diameter and a length of about 34 inches. A wire screen near the bottom of each column served as a support for about 24 inches of sand, topped by a one-inch layer of 4 mm pea gravel to distribute the intermittent dosage and minimize scum formation. Ten of the initial columns were packed with a medium-sized silica sand having a geometric mean size (M_g) of 0.56 mm and a geometric standard deviation (S_g) of 1.2. The other ten columns had a finer sand with an M_g of 0.12 mm and a S_g of 1.14, on top of about an inch of the medium sand in contact with the wire screen.

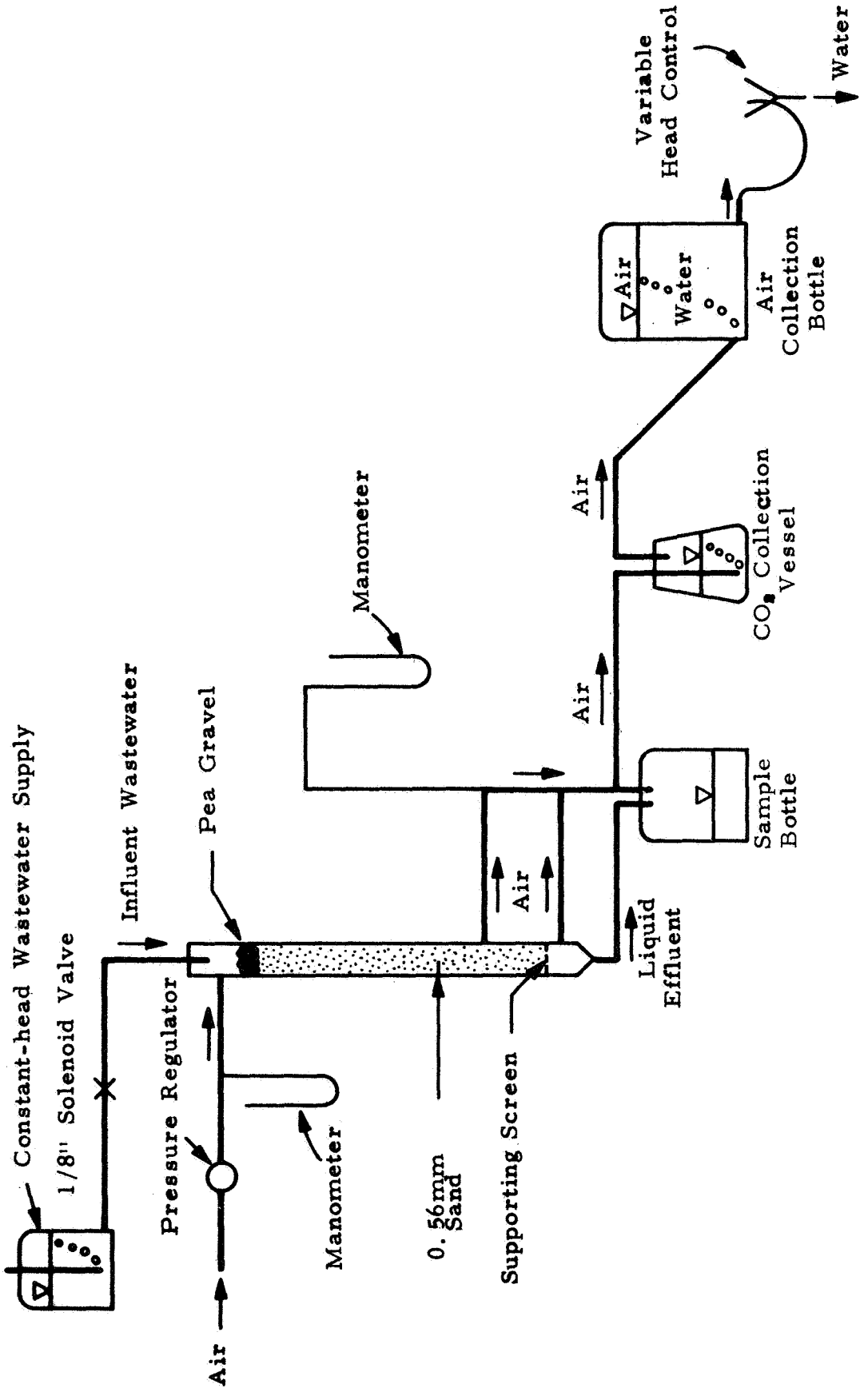


Figure 5.1--SCHEMATIC DIAGRAM OF INTERMITTENT SAND FILTRATION APPARATUS.

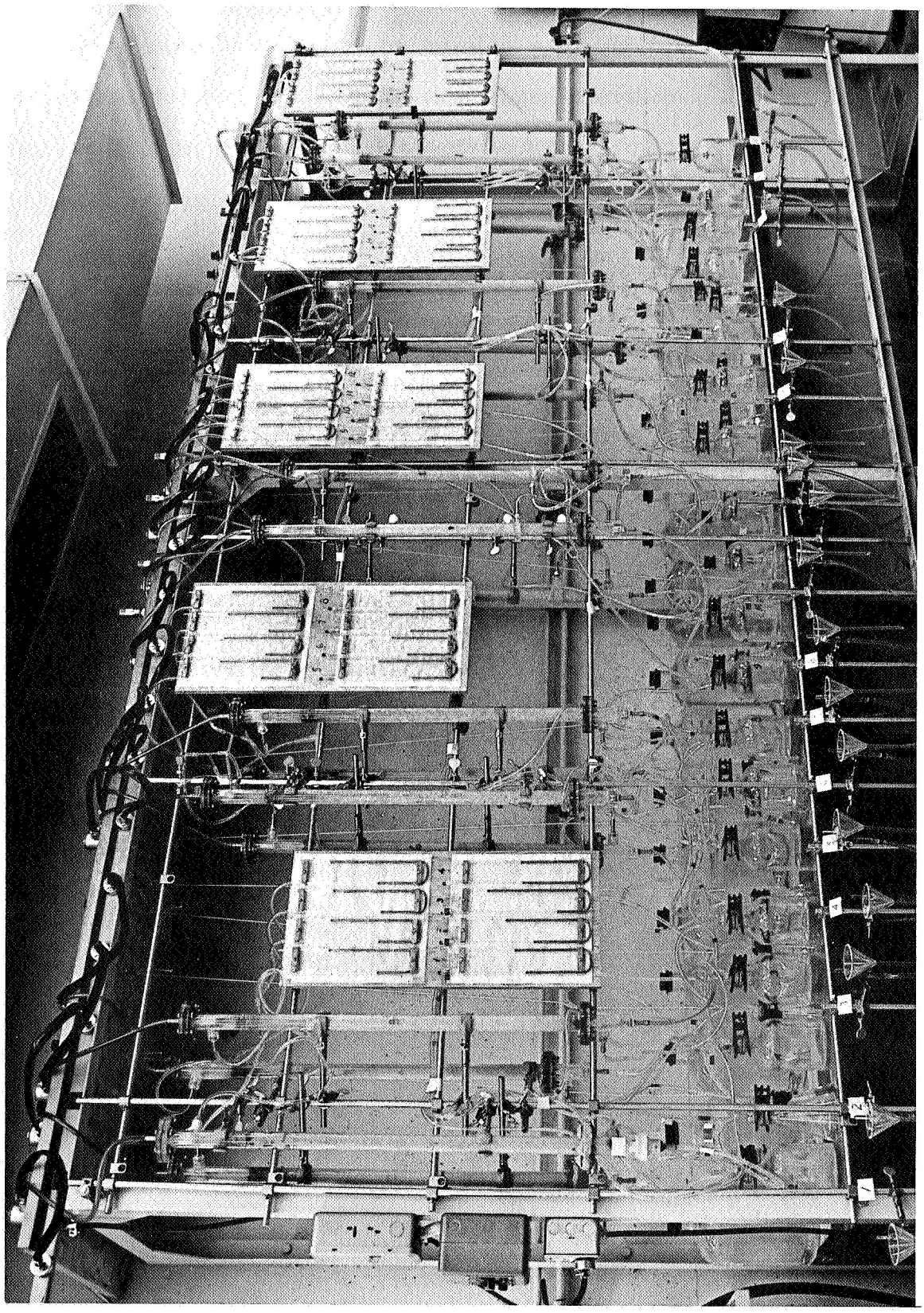


Fig. 5.2 VIEW OF INITIAL SAND COLUMNS

The liquid fed to a column was held in a glass supply bottle with a constant head controlled by the bubbler device, as shown by Fig. 5.1. It was dosed intermittently through a 1/8-inch solenoid valve controlled by a timer that could be actuated over a wide frequency range, i.e. once every 15 minutes to once per day. The duration of the aqueous flow to the column could be controlled from almost zero to a maximum of 15 seconds. The volume of each dosage was governed by the time the solenoid valve was open and by the hydraulic head from the supply bottle, which could be raised or lowered as necessary. Initially each column received about 12 ml of liquid in about two seconds once every two hours (or 12 times per day), making a total hydraulic load of about 144 ml or about 28 cm^3 per cm^2 per day, i.e. 28 cm per day. This rate corresponds approximately to that which can be achieved when settled municipal sewage is applied intermittently to sand beds. With the laboratory columns, however, it proved difficult to regulate the dosing mechanism carefully enough to provide equal hydraulic loading rates on all filters, but the total loading for each day was recorded in the volume of collected percolate. Later in the project, this liquid feeding system was modified, as described in Section 5.02.

Inasmuch as preliminary computations indicated that the oxygen requirement for complete nitrification of 150 ml of full-strength urine would be in the order of 4,800 ml of pure oxygen

at STP, corresponding to about 24,000 ml per day of air, it was evident that simple atmospheric diffusion into the sand column would probably not be adequate to keep the system aerobic. Provision was made, therefore, to apply compressed air or oxygen in such a way as to create a forced draft through the column. This was not a simple task in view of the fact that (a) the surface of the sand was flooded intermittently and (b) a capillary fringe formed at the bottom of the column. It was accomplished by means of the system shown in Fig. 5.1 consisting of a pressure regulator, influent and effluent manometers for measuring pressure differentials, and provision for taking the air from the column by means of a manifold located above the capillary fringe. Arrangements were also made to bubble the effluent air through a bottle containing a known quantity of KOH or BaOH to strip out the CO_2, followed by a large displacement bottle in which the total remaining volume of air could be measured. The differential head on the compressed air flow was controlled by the pressure regulator at the influent end and by the variable head control of the water displaced from the air collection bottle.

The capillary fringe at the bottom of each column varied from one to four inches in depth, depending on the grain size and the method of packing. In order to obtain a flow of air through this saturated zone, it was necessary to maintain a very high pressure differential. The resultant large volume of air was difficult to collect and measure, and it was far in excess of the

need for oxygen. To eliminate this difficulty, an air outlet was placed six inches above the supporting screen and well above the capillary fringe. This arrangement permitted an adequate air flow through the top 18 inches of sand with only a slight differential in pressure.

Difficulties were experienced with the air flow system when natural urine was fed to the columns. At that time, the increase in biological growths in the upper few inches of the sand was so excessive that air flow was blocked and anaerobic conditions developed. Anaerobiosis, in turn, increased the clogging. As a result, the influent urine ponded on the surface and filled the small air cavity above the sand. This problem could be ameliorated only by decreasing the total amount and/or the strength of the daily urine dosage, by frequent scarifying of the upper two or three inches of sand, by changing the grain size of sand in the columns, or by a combination of these palliatives.

Several of the columns gave evidence of crazing or cracking, but there was no indication of air leakage, based on soap films. In addition, algae developed in some of the columns since the clear lucite permitted penetration of some light. To mitigate both of these problems, the columns were wrapped with black photographic paper.

5.02 Modifications to the Sand Columns

Midway through the experimentation it became necessary to reconstruct the laboratory columns. With the initial system, close control of the liquid feed rate in the range of 10 to 20 ml

per intermittent dose was difficult to achieve. The variation of flow between successive dosages was often as high as 50 percent. Part of the control problem was attributable to pressure fluctuations between the feed supply bottle and the air cavity when the column was being dosed.

To improve control of the dosage volume, a fluid restrictor was placed between the solenoid valve and the column, as shown in Fig. 5.3. The feed bottle was also raised to a higher position above the column. The first restrictor used consisted of a glass tubing (8 mm I. D.) filled with sand; but because of biological clogging, that device was replaced by a short piece of capillary tubing (1.0 mm I. D.). The capillary restrictor proved to be successful and in conjunction with a timer span of 15 seconds it provided an excellent means for controlling the dosage volume over the ranges desired. The problem of pressure fluctuation during dosing was simply resolved by installing an equalizer line between the feed bottle and the air line to the column (see Figures 5.3, 5.4, and 5.5).

Modifications to the air-control system involved the insertion of needle valves between a pressure-regulated header and the individual columns. The air pressure in the header was controlled by a common pressure regulator but the discharge to each column was adjusted by the individual needle valves. With the needle valve in this new position and by maintaining a pressure drop of about four inches of water across the valve,

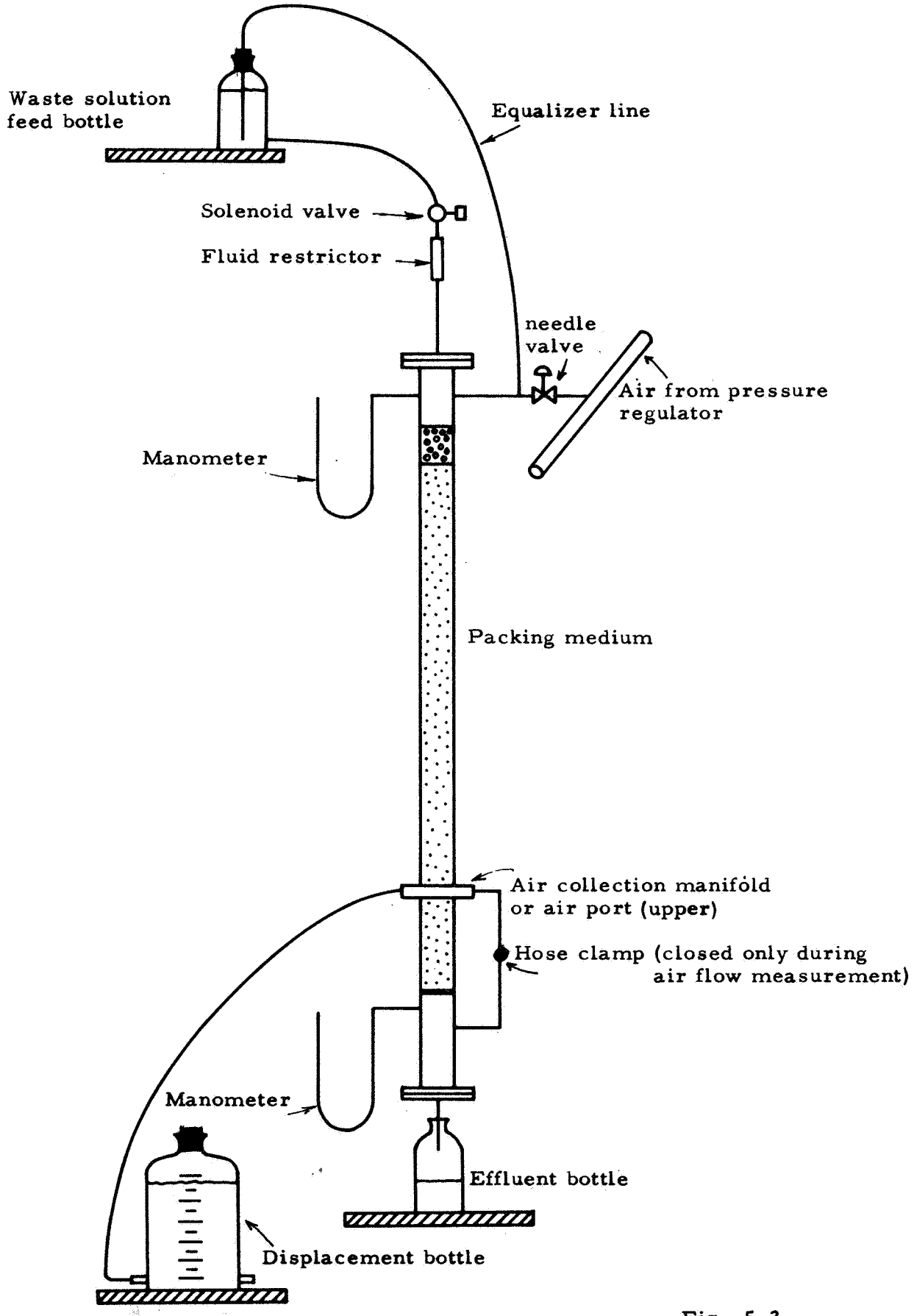


Fig. 5.3
Schematic Diagram of
Modified Column

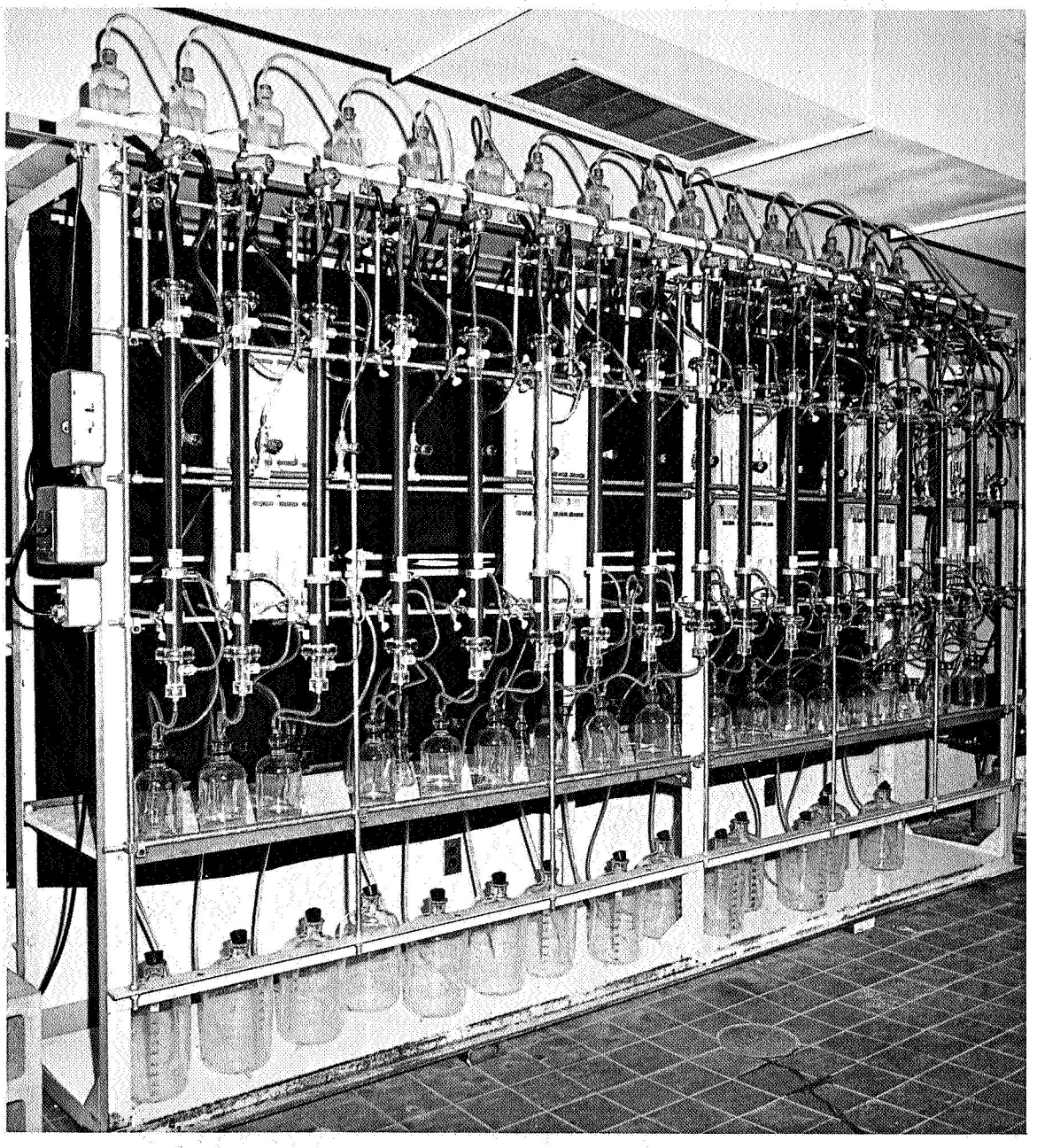


Fig. 5.4 VIEW OF MODIFIED SAND COLUMNS

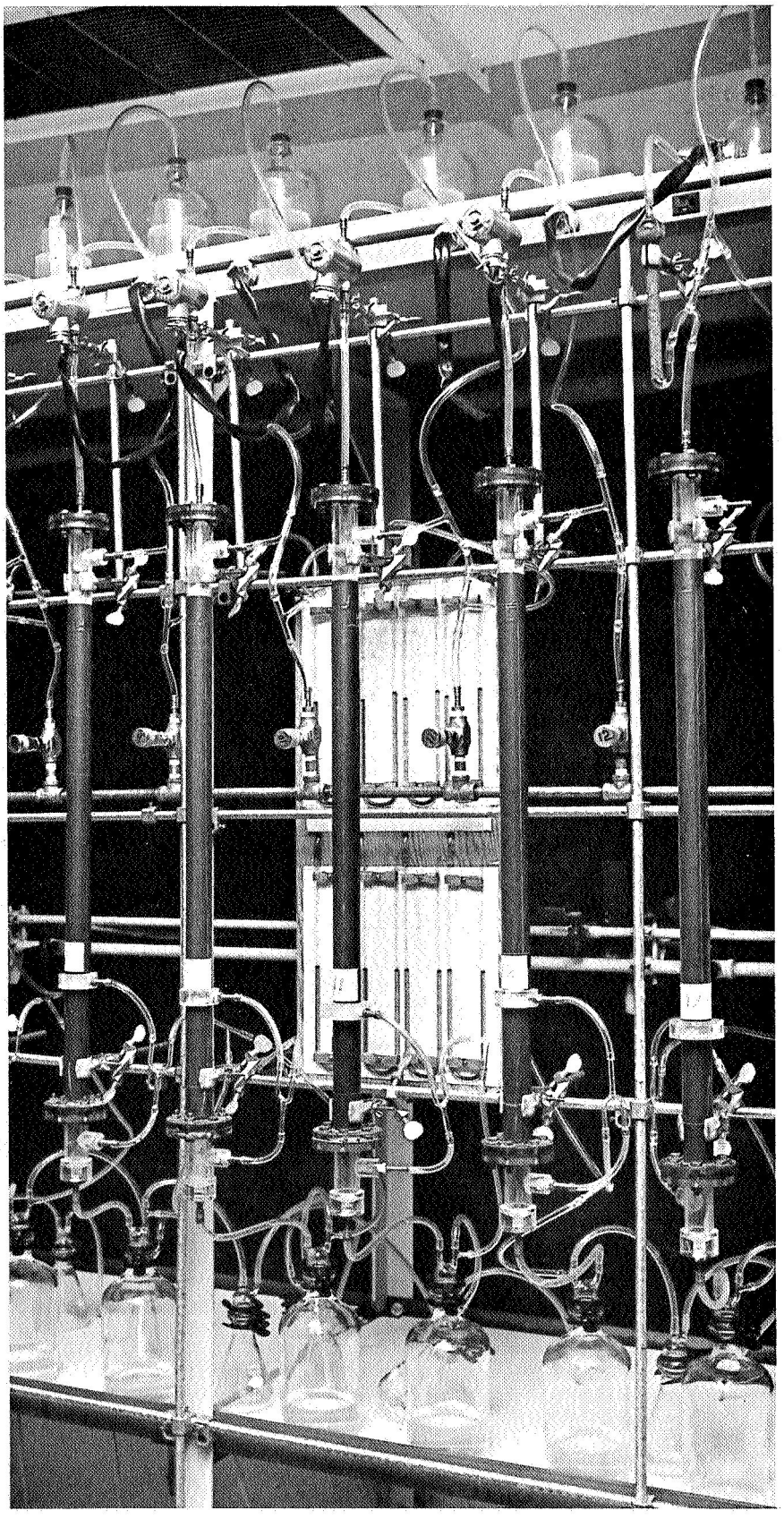


Fig. 5.5 CLOSER VIEW OF MODIFIED SAND COLUMNS

the air rate could be controlled to as low as 0.2 liters per hour. The four-inch pressure differential across the needle valve was necessary to accommodate increases in impedance arising from biological growths in the column. The upper manometer, attached to the air cavity, proved to be a useful index of the status of biological growth.

The air displacement bottle was used only for calibrating the adjustment of the needle valve periodically. When not so needed, the air displacement bottle was disconnected. Furthermore, the carbon-dioxide trap was discontinued when it became apparent at this stage of the project that carbon metabolism was of minor interest compared with the difficulties in achieving nitrification of ammonia.

5.03 Further Modifications

It appeared prudent during the latter part of the experimentation to determine if nitrification would be favored by longer retention periods and deeper columns. The short 30-inch columns were joined together in series to form the 60-inch columns shown at the right side of Figure 5.6. In order to provide an instantaneous check on the air flow to each such column, an Erlenmeyer flask (bubbler) was placed in the air line between the needle valve and the column, as shown in Figure 5.7. The flask was filled partially with water. A 2-mm I.D. capillary tubing was placed inside and approximately 2-3 cm below the air-water interface. By counting the bubbles per minute and knowing

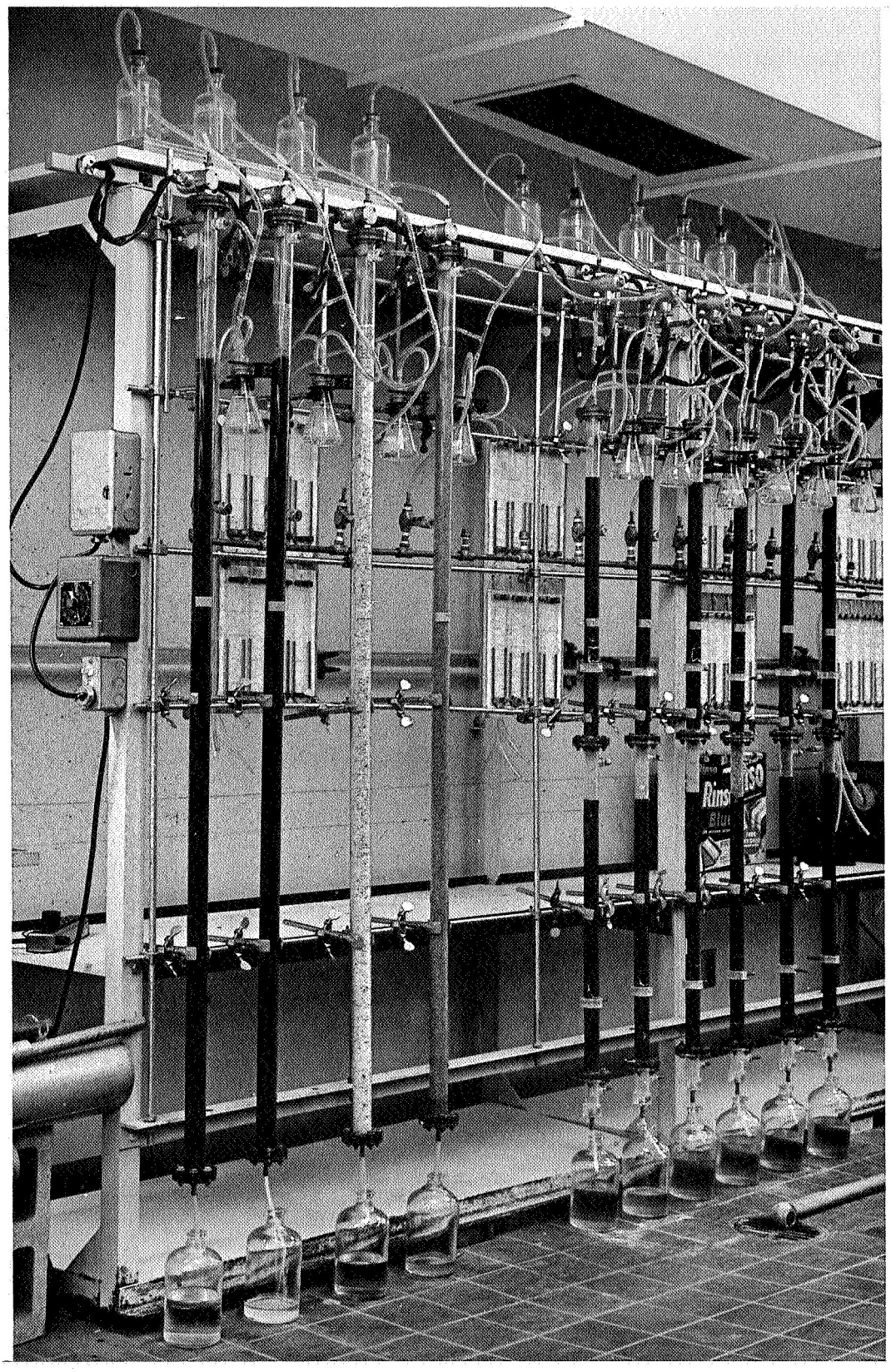


Fig. 5.6 ACTIVATED CARBON AND CALCIUM CARBONATE COLUMNS

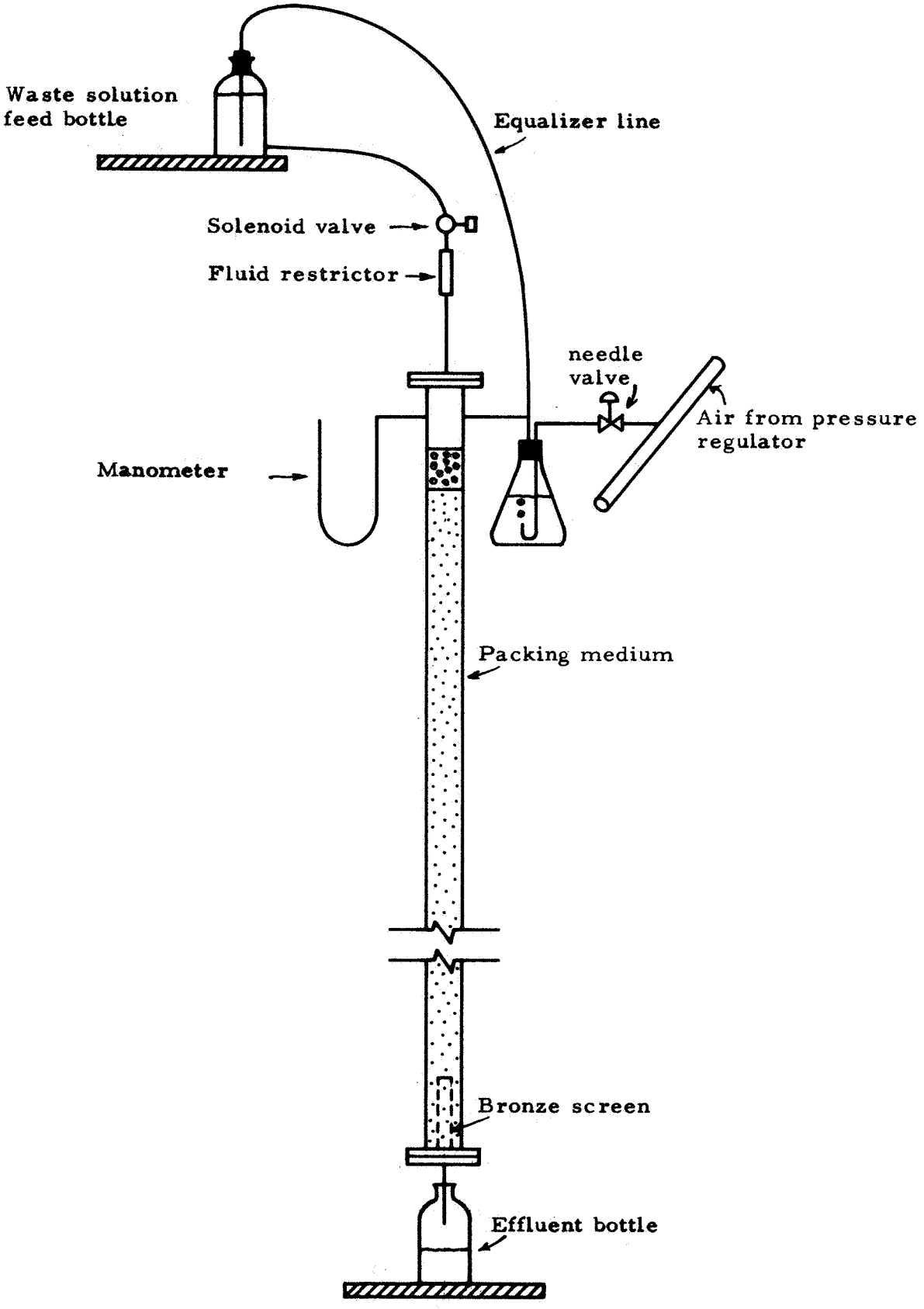


Fig. 5.7
Schematic Diagram of
Modified Air Control System

the volume of each bubble, the air flow rate could be measured quite accurately and controlled carefully by the needle valve. Occasional calibration checks on the bubble flow rate were made with the air displacement bottle.

5.04 Columns for Activated Carbon and $CaCO_3$

Near the end of the project, four new columns were constructed to contain activated carbon or calcium carbonate. They were made of lucite tubing, 1.25 inches in diameter and six feet long. These columns are illustrated at the left side of Fig. 5.6. A new system for removing air and liquid effluent was incorporated into the design in order to reduce fabrication costs. In lieu of the doughnut-shaped air-collection manifold six inches above the horizontal effluent screen, a bronze tubular screen, 3/4 inch in diameter and 6 inches long, was secured to the bottom flange and inserted into the lower end of the column, as shown in Figure 5.7. This new column design was extremely simple and less subject to air leakage--a problem that invariably arises when gluing lucite surfaces such as the air collection manifold.

Two of the four new columns were packed with granular activated carbon having an M_g of approximately 0.56 mm. This medium was chosen because of its high adsorptive capacity, on the hypothesis that increased adsorption would favor the retention of ammonia in a column sufficiently long to allow nitrification to take place. The other two columns were packed with a mixture

of coarse $CaCO_3$ particles (about 4 mm diameter) and 0.56 mm sand. The $CaCO_3$ was used for its buffering capacity, in anticipation that nitrification would be favored if the pH value could be maintained in the 7.5-8.5 range.

5.05 Analytical Procedures

In the early phases of the project, the analyses of the influent to and effluent from the laboratory columns were intended to cover all of the following parameters:

Kjeldahl nitrogen (organic N + ammonia)

Ammonia

*Urea

*Creatinine

Nitrites

Nitrates

**Total solids (residue on evaporation)

**Chlorides

**Carbon dioxide

Chemical oxygen demand (COD)

pH value

As the program developed it became evident that the tests marked ** above were not especially relevant to the mission of trying to achieve nitrification and other biochemical stabilization. In view of this fact and the manpower and budget limitations, these tests were discontinued early in the program.

When it became evident that urea and creatinine in the

applied urine were being hydrolyzed rapidly and completely to ammonia, the tests marked * were also terminated. Although trace quantities of creatinine were occasionally found in the effluent, urea never appeared in the percolate. Consequently, all nitrogen balances were based on Kjeldahl nitrogen, ammonia, nitrates, and nitrites.

The test methods employed to determine the concentration of the respective nitrogen compounds were found in some instances to be highly inaccurate. This observation was particularly true with regard to the spectrophotometric methods for determining nitrite and nitrate. Interferences from the colored components (bile pigment) contained in natural urine required that alternative methods be sought. After experimenting with various methods and techniques, success was finally realized. A brief discussion of each of the methods employed to determine the nitrite and nitrate concentrations as well as all of the other methods used to measure the remaining compounds and biological parameters is presented below.

Chemical oxygen demand (COD)

The method selected for measuring the chemical oxygen demand, and most of the other methods used in this project, are described in Standard Methods (34). To eliminate the interference from nitrite (oxidized to nitrate by the potassium dichromate) required the addition of relatively large quantities of sulfamic acid (1300 mg sulfamic acid per liter of dichromate).

Nitrite

The nitrite concentration was obtained by a somewhat novel but accurate method that consisted of performing the normal COD but without the addition of sulfamic acid. Without the preferential oxidation by the sulfamic acid, the nitrite was quantitatively oxidized by the dichromate to nitrate. The nitrite concentration was subsequently calculated by subtracting the true COD from this pseudo-COD and multiplying the difference by the molecular weight ratio (M.W. of N/M.W. of O) to obtain the nitrate nitrogen equivalent. Because the effluent was essentially free of organic nitrogen compounds, some of which can reduce nitrite to nitrogen, no interferences were observed.

Nitrate

The nitrate concentration was determined by a nitrate electrode in conjunction with an EMF or pH meter. Both the electrode and EMF meter were manufactured by Orion Research, Inc. The electrode was reasonably specific and consequently corrections for interferences from the bicarbonate and nitrite ions were not necessary unless a high degree of accuracy was desired.

Unfortunately, the precision of this electrode like any other electrode was strongly dependent on the ionic strength of the solution. Errors introduced by variations in ionic strength resulting from the biological oxidation of the urine were removed by careful preparation of the calibrating solution. Increased accuracy with the electrode was obtainable by diluting both the

calibrating and sample solution 10 to 1. The electrode was less sensitive to small variations in ionic strength of these solutions if the concentration was reduced ten fold.

Ammonia

The ammonia content of the effluent was determined by the distillation method. The procedure of first buffering the sample with KH_2PO_4 and K_2HPO_4 to maintain the pH at 7.4 proved to be unsatisfactory. During the distillation step as much as 80 mg N per liter of ammonia was lost or, more precisely, unaccounted for. In short, both accurancy and repeatability were extremely poor. A much more accurate procedure was worked out that consisted of first boiling the sample with a dilute H_2SO_4 solution and allowing the nitrous acid to escape. After this step the sample was made alkaline by the addition of NaOH and the ammonia was absorbed into a boric acid solution. The repeatability of this test was very satisfactory (± 2 percent). The results of this test were periodically checked against those obtained from the total Kjeldahl nitrogen test (explained below) in order to assure that the sample contained no organic nitrogen and that no interferences (volatile components other than ammonia) were affecting the ammonia test just described. Deviations between the results of these two tests were very small.

Nitrogen

The Kjeldahl acid digestion method was employed to determine the total nitrogen of the influent and occasionally, as stated above, the ammonia content of the effluent. Since no

nitrogen compounds in natural urine are considered highly refractory, no inorganic salts (e.g. K_2SO_4) were used to raise the boiling point of the digest. Only H_2SO_4 and a small quantity of $CuSO_4$ (catalyst) were used in formulating the digest. Prolonged digestion periods were not necessary, as the solution cleared very rapidly after the H_2SO_4 began to boil. After cooling, the solution was made alkaline and the ammonia was absorbed into boric acid and subsequently titrated with 0.02N H_2SO_4.

Hydrogen ion concentration (pH)

The pH value was determined by means of a standard Beckman meter. The pH was a crude but rapid method for ascertaining the biological status of the columns during the ripening period. If nitrification was weak, no significant difference between the influent and effluent pH was observed. Conversely, when the essential nitrifiers were present and oxidizing ammonia to nitrite and nitrate, a noticeable drop in pH value was observed.

CHAPTER VI
EXPERIMENTAL RESULTS

6.01 Settled Municipal Sewage.

Primary effluent (settled sewage) from the Whittier Narrows Water Reclamation Plant was used to ripen the initial sand columns. It was filtered through glass wool to remove suspended solids that might cause excessive clogging of the sand.

The first column was placed in operation in August 1965 in order to check the design before the other 19 columns were constructed. It received intermittent doses of sewage totalling 100 to 150 ml per day, for a hydraulic loading of about 20 to 30 cm per day. Almost immediately the removal of COD was highly effective, largely as a result of physical adsorption combined later with biochemical oxidation of adsorbed carbonaceous matter. The influent COD averaged about 225 mg per liter while the effluent contained about 35 mg per liter, corresponding to about 85 percent removal.

The oxidation of organic and ammonia nitrogen to nitrates, however, did not begin to occur for about 40 days. By mid-October 1965, approximately two-thirds of the applied nitrogen was being converted to nitrates, but this efficiency did not improve through most of November. This initial filtration occurred with natural diffusion of air into the column, which was open to the atmosphere. On the supposition that the nitrifying bacteria were not getting sufficient oxygen, especially in the deeper parts of the sand, the column was then connected to a pure oxygen supply for about two months.

Instead of improving, however, nitrification deteriorated. A review of soil-science literature indicated that high oxygen tensions appear to have a detrimental effect on nitrifying microorganisms. The pure oxygen supply was removed in mid-January 1966 and the column was connected to a compressed-air line. Nitrification improved thereafter but seldom exceeded 60 percent.

Meanwhile, 19 additional columns were constructed, connected to the compressed air line, and ripened for five months with settled municipal sewage. The results of tests in February and March 1966 for 16 columns are presented in Table 6.1. The removal of COD averaged 86 percent on these days of analyses but the oxidation of nitrogen to nitrates was only 50 percent effective. It is interesting to note that the poorest performance, both for COD and nitrification, occurred with column No. 1, which had been subjected to the shock effect of the pure oxygen atmosphere. Apparently, it had not recovered fully.

Since the purpose of these preliminary tests was not to assess the columns' efficiency in the treatment of municipal sewage but rather to check out the design and ripen the columns with nitrifying organisms, no attempt was made to improve the degree of nitrification by altering the size or type of media or modifying the dosing cycle or the hydraulic loading rate. As noted in Chapter V, the finer sand with a M_g of 0.12 mm had proved unsatisfactory and was replaced by a medium silica sand. In all probability, the extent of nitrification could have been improved by decreasing the hydraulic load such that the daily volume of water applied was equal or

Table 6.1
BIOLOGICAL STABILIZATION OF SETTLED MUNICIPAL SEWAGE

Column	Effluent COD, mg per liter				Effluent nitrate, mg N per liter			
	2/10/66	2/28/66	3/7/66	Average	2/10/66	2/28/66	3/7/66	Average
# 1	43	32	47	41	5	14	9	9
# 2	29	-	30	30	10	-	21	16
# 3	35	27	38	33	10	25	15	17
# 4	28	30	29	29	15	12	24	17
# 5	30	23	33	29	18	24	22	21
# 6	34	36	35	35	16	10	7	11
# 7	39	25	33	32	16	24	22	21
# 8	38	24	32	32	17	22	23	20
# 9	40	33	35	35	5	25	10	13
#10	48	30	42	38	13	19	16	16
#11	26	24	42	31	17	22	24	21
#12	22	20	34	25	-	10	30	20
#13	40	29	45	38	10	22	10	14
#14	40	35	45	40	16	22	19	19
#15	32	31	24	29	-	10	17	14
#16	30	22	31	28	7	25	25	19
				Average---- 33				Average--17

Average Influent COD-- 237

% Removal----------86%

Average Influent Nitrogen Content*-----34

% Nitrogen Oxidized to nitrate----------50%

Column Pack Medium - Silica Sand, M_g = .56 mm, S_g = 1.2

Dosage Frequency 12 cycles/day Dosage Volume 12 ml/dose

*Total Kjeldahl nitrogen (organic and ammonia). Influent had no measurable nitrates or nitrites.

less than the volume of pellicular and interstitial water in the column, as explained in Appendix B. Experience with intermittent sand filtration of settled municipal sewage for almost a century has shown that essentially complete nitrification can be achieved when the organic and hydraulic loads are sufficiently low.

6.02 Natural and Synthetic Urine.

The results, experiences, and difficulties associated with the application to the well-ripened columns of natural or synthetic urine in dilutions up to 20:1 are described in Sections 4.03 and 4.04 of this report and will not be repeated here. Attempts to achieve any significant degree of nitrification for urine diluted with distilled water were largely unsuccessful despite all of the modifications described in Chapter IV. The difficulties experienced with the dosing mechanism and clogging are recounted in Chapters IV and V. Finally, near the end of 1967, the columns were rebuilt and the dosing equipment was improved sufficiently to enable the project engineer to embark on a new set of experiments.

The urine used in this phase of the project came from one man only (the project engineer). Although no significant changes in his urine composition had been observed for several months, it was desirable to avoid any daily fluctuations in strength and to reduce the analytical work. Hence, the urine was stored in a four-liter bottle in a cold room at about 3.5°C to minimize decomposition and the bottle was kept full by daily contributions. The mixed urine was withdrawn daily, diluted to the proper concentrations, and placed

immediately in the feed bottles. This mixture had a Kjeldahl nitrogen concentration of about 7600 mg/l and a COD of about 7500 mg/l. The columns were operated for four months on the same feed before the experiments described hereinafter were conducted.

6.03 Oxygen Demand of Natural Urine.

Based on a Kjeldahl nitrogen concentration of 7600 mg per liter, a COD of 7500 mg/l, and a maximum dosage of 200 ml per day, the total oxygen requirement for complete nitrification and for complete oxidation of the carbonaceous matter would be as follows:

Total nitrogen dose = 7600 x 0.2 = 1520 mg/day

Oxygen requirement for nitrification = 64/14 x 1520 = 6950 mg/day

Carbonaceous oxygen requirement = 0.2 x 7500 = 1500 mg/day

Total oxygen requirement = 6950 + 1500 = 8450 mg day

Oxygen volume at STP = 8450 x 22.4/32.0 = 5900 ml/day

Equivalent volume of air = 5900 ÷ 0.21 = 28,200 ml/day

For a 10-percent urine dilution, the air requirement is approximately 2820 ml per day, and for a 20-percent dilution, about 5640 ml per day. In the experiments described hereinafter, the air flow rate was maintained at 7400 ml per day for all tests; hence the supply of oxygen should not have been a limiting factor in preventing complete nitrification at the dilutions used.

Yet, as described hereinafter, high concentrations of nitrite rather than nitrate, were encountered in several experiments, which leads one to suspect that the supply of oxygen may have been inadequate in certain parts of the column, i.e. the forced air supply

may have short-circuited or channeled through the column, leaving many pockets deficient in oxygen. This point is discussed in more detail for individual experiments.

6.04 Silica Sand Columns.

Seven columns of silica sand were tested under the following conditions:

Column Number	Percent Dilution	Size of Sand	Hydraulic Rate, cm per day
5	10	Medium	19.2
7	10	Medium	19.2
4	10	Coarse	22.7
8	10	Coarse	19.2
9	20	Coarse	19.2
10	5	Coarse	19.2
6	10*	Medium	19.2

*Column No. 6 received 10-percent synthetic urine while the other six columns were dosed with natural urine in the indicated dilutions. The results of these experiments are presented in Tables 6.2 to 6.8 inclusive.

The conditions under which columns No. 5 and No. 7 operated were similar (medium sand, 10-percent dilution) and in many ways the results were comparable, e.g. about the same percentage of unaccounted-for nitrogen (3.5-3.7%) and about the same percentage of ammonia-N in the effluent (44.8-44.9%). The distribution of oxidized nitrogen, however, between nitrites and nitrates was quite different. For some reason or other, the Nitrobacter organisms in column No. 5 appear to have been more inhibited than those in column No. 7, despite the fact that pH values of the effluent from the latter column were lower.

Table 6.2

NITRIFICATION OF NATURAL URINE AT 10:1 DILUTION
COLUMN NO. 5---SILICA SAND, MEDIUM

Physical data.

Diameter of column ---------- 1.0 inch
Depth of media ---------- 54 inches
Media:-----silica sand
M_g ---------- 0.56mm
S_g ---------- 1.2
Hydraulic loading rate---------100 ml per day = 19.2 cm per day
Dosing frequency---------- 4 times per day
Quantity per dose ---------- 25 ml
Air flow rate ----------7400 ml per day

Strength of natural urine at 10:1 dilution.

Kjeldahl nitrogen---------- 760 mg N per liter
pH value----------5.8

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	6.4	6.1	5.7	6.1
Ammonia, mg N per liter	325	330	340	332
Nitrite, " " " "	325	400	365	367
Nitrate, " " " "	75	20	40	45
Total N " " " "	725	750	745	740
Unaccounted for N, %	4.6	1.3	2.0	3.7
Ammonia, % of effluent N	44.8	44.0	45.6	44.9
Effluent ammonia, % of input N	42.7	43.4	44.7	43.6

Table 6.3

NITRIFICATION OF NATURAL URINE AT 10:1 DILUTION
COLUMN NO. 7---SILICA SAND, MEDIUM

Physical data.

Diameter of column ---------- 1.0 inch
Depth of media ---------- 54 inches
Media: ---------- silica sand
M_g ---------- 0.56 mm
S_g ---------- 1.2
Hydraulic loading rate ---------- 100 ml per day = 19.2 cm per day
Dosing frequency ---------- 4 times per day
Quantity per dose ---------- 25 ml
Air flow rate ---------- 7400 ml per day

Strength of natural urine at 10:1 dilution.

Kjeldahl nitrogen ---------- 760 mg N per liter
pH value ---------- 5.8

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	5.4	5.2	6.0	5.5
Ammonia, mg N per liter	335	330	320	328
Nitrite, " " " "	280	135	135	183
Nitrate, " " " "	85	310	270	222
Total N, " " " "	700	775	725	733
Unaccounted for N, %	7.9	(-2.0)	4.6	3.5
Ammonia, % of effluent N	48.0	42.5	44.1	44.8
Effluent ammonia, % of input N	44.1	43.5	42.1	43.1

Table 6.4

NITRIFICATION OF NATURAL URINE AT 10:1 DILUTION
COLUMN NO. 4---SILICA SAND, COARSE

Physical data.

Diameter of column	1.25	inches
Depth of media	60	inches
Media:	silica sand	
Grain size max. (passed 12 mesh sieve)	1.40	mm
Grain size min. (retained on 14 mesh sieve	1.17	mm
Hydraulic loading rate	180	ml per day = 22.7 cm per day
Dosing frequency	4	times per day
Quantity per dose	45	ml
Air flow rate	7400	ml per day

Strength of natural urine at 10:1 dilution.

Kjeldahl nitrogen	760	mg N per liter
pH value	5.8	

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	5.5	6.1	6.1	5.9
Ammonia, mg N per liter	340	320	325	328
Nitrite, " " " "	380	370	360	370
Nitrate, " " " "	40	50	80	57
Total N, " " " "	760	740	765	755
Unaccounted for N, %	0.0	2.6	(-0.7)	0.6
Ammonia, % of effluent N	44.8	43.2	42.5	43.5
Effluent ammonia, % of input N	44.8	42.1	42.8	43.1

Table 6.5

NITRIFICATION OF NATURAL URINE AT 10:1 DILUTION
COLUMN NO. 8---SILICA SAND, COARSE

Physical data.

Diameter of column	1.0	inch
Depth of media	54	inches
Media:	silica sand	
Grain size max. (passed 10 mesh sieve)	1.6	mm
Grain size min. (retained on 12 mesh sieve)	1.4	mm
Hydraulic loading rate	100	ml per day = 19.2 cm per day
Dosing frequency	4	times per day
Quantity per dose	25	ml
Air flow rate	7400	ml per day

Strength of natural urine at 10:1 dilution

Kjeldahl nitrogen	760 mg N per liter
pH value	5.8

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	6.0	5.9	5.6	5.8
Ammonia, mg N per liter	370	335	300	335
Nitrite, " " " "	300	140	155	198
Nitrate, " " " "	70	260	300	210
Total N, " " " "	740	735	755	743
Unaccounted for N, %	2.6	3.3	0.7	2.2
Ammonia, % of effluent N	50.0	45.5	39.8	45.1
Effluent ammonia, % of input N	48.6	44.0	39.5	44.0

Table 6.6

NITRIFICATION OF NATURAL URINE AT 5:1 DILUTION
COLUMN NO. 9---SILICA SAND, COARSE

Physical data.

Diameter of column	1.0	inch
Depth of media	54	inches
Media:	silica sand	
Grain size max (passed 10 mesh sieve)	1.6	mm
Grain size min (retained on 12 mesh sieve)	1.4	mm
Hydraulic loading rate	100	ml per day = 19.2 cm per day
Dosing frequency	4	times per day
Quantity per dose	25	ml
Air flow rate	7400	ml per day

Strength of natural urine at 5:1 dilution.

Kjeldahl nitrogen	1520	mg N per liter
pH value	5.8	

Effluent analyses

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	6.8	7.4	6.3	6.8
Ammonia, mg N per liter	700	740	600	680
Nitrite, " " " "	570	530	620	573
Nitrate, " " " "	30	140	60	77
Total N, " " " "	1300	1410	1280	1330
Unaccounted for N, %	14.5	7.2	15.8	12.5
Ammonia, % of effluent N	53.9	52.5	46.8	51.0
Effluent ammonia, % of input N	46.0	48.6	39.5	44.8

Table 6.7

NITRIFICATION OF NATURAL URINE AT 20:1 DILUTION
COLUMN NO. 10---SILICA SAND, COARSE

Physical data.

Diameter of column	1.0	inch
Depth of media	54	inches
Media:	silica sand	
Grain size max. (passed 10 mesh sieve)	1.6	mm
Grain size min. (retained on 12 mesh sieve)	1.4	mm
Hydraulic loading rate	100	ml per day = 19.2 cm per day
Dosing frequency	4	times per day
Quantity per dose	25	ml
Air flow rate	7400	ml per day

Strength of natural urine at 20:1 dilution.

Kjeldahl nitrogen	380	mg N per liter
pH value	5.8	

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	6.0	5.8	6.3	6.0
Ammonia, mg N per liter	180	200	160	180
Nitrite, " " " "	0	0	50	17
Nitrate, " " " "	200	205	180	195
Total N, " " " "	380	405	390	392
Unaccounted for N, %	0.0	(-6.6)	(-2.6)	(-3.2)
Ammonia, % of effluent N	47.5	49.4	41.0	46.0
Effluent ammonia, % of input N	47.3	52.6	42.1	47.3

Table 6.8

NITRIFICATION OF SYNTHETIC URINE AT 10:1 DILUTION
COLUMN NO. 6---SILICA SAND, MEDIUM

Physical data.

Diameter of column	1.0	inch
Depth of media	54	inches
Media:	silica sand	
M_g	0.56	mm
S_g	1.2	mm
Hydraulic loading rate	100	ml per day = 19.2 cm per day
Dosing frequency	4	times per day
Quantity per dose	25	ml
Air flow rate	7400	ml per day

Strength of synthetic urine at 10:1 dilution.

Kjeldahl nitrogen ---------- 870 mg N per liter

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	5.8	6.1	5.2	5.7
Ammonia, mg N per liter	400	400	410	403
Nitrite, " " " "	250	290	220	253
Nitrate, " " " "	200	150	240	197
Total N, " " " "	850	840	870	853
Unaccounted for N, %	2.3	3.4	0.0	2.0
Ammonia, % of effluent N	47.0	47.6	47.1	47.2
Effluent ammonia, % of input N	46.0	46.0	47.1	46.4

Tables 6.4 and 6.5 indicate that the use of a coarser sand had little, if any, effect on nitrification, as measured by the percentage of ammonia in the percolate. Again, there was a marked but inexplicable difference in the distribution of oxidized nitrogen between nitrates and nitrites, even when pH values were comparable.

With stronger urine (20 percent, Table 6.6) the conversion of ammonia nitrogen to nitrites was still quite good, but the final oxidation to nitrate was minimal, despite almost neutral pH values. The only logical explanations for the poor performance by Nitrobacter organisms would be that (a) insufficient oxygen was available, as a result of short-circuiting of the forced air draft (b) the column wasn't deep enough to provide time for the Nitrobacter to act after the Nitrosomonas had converted ammonia to nitrites, or (c) despite previous ripening, the column was not well-seeded with Nitrobacter.

With weaker urine (5 percent, Table 6.7) the destruction of ammonia nitrogen was not any more complete than at 10 percent or 20 percent dilutions, but the conversion of nitrite to nitrate was much more effective. This column apparently was well seeded and had adequate oxygen.

Synthetic urine at 10-percent dilution (see Table 6.8) appears to have responded to intermittent filtration through medium silica sand in much the same way as did natural urine.

The nitrogen balances for the silica sand columns, as noted by the unaccounted-for nitrogen, were reasonably good and generally within the accuracy of the analytical methods. In almost all experiments, the total nitrogen in the percolate was slightly less

than that in the nitrogen input, and in a few tests, the output was greater than the input. These results are reasonable when one considers that some of the nitrogen is stored temporily in the column, especially by heterotrophic bacteria, and later released. Hence, one cannot expect to achieve a perfect nitrogen balance every day. Furthermore, some of the ammonia may be lost by gas transfer to the atmosphere, for indeed the odor of ammonia was frequently noticeable near the columns.

The loss of nitrogen was especially evident for the 20-percent dilution (Table 6.6). When coupled with the poor conversion of nitrite to nitrate in this column, this loss suggests that portions of the filter may have been anaerobic and that some denitrification (to nitrogen gas) may have been occurring.

Complete nitrification to nitrates, or only to nitrite, followed by denitrification and the production of nitrogen gas might prove to be advantageous for space travel. This supply of nitrogen gas could be used as a replacement for nitrogen lost by slight leakages through the space vehicle walls to the outside vacuum.

It is probable that stratification of reactions in the columns may have been a most significant factor in determining the degree of nitrification. As described in Sections 3.01 and 3.02 of this report, the hydrolysis of urea, creatinine, etc. to ammonium ion occurs quite rapidly and completely in a non-sterile environment. This reaction probably takes place in the upper few inches of the filter, but it may be delayed to greater depths, although numerous tests showed that even in a 24-inch deep filter, hydrolysis of urea was

virtually 100 percent complete. The hydrolysis is accompanied by a rise in pH from about 5.8 in urine to about 8.8 in the hydrolyzed state. At pH 8.8, some of the ammonium ion will be converted to undissociated ammonium hydroxide, which is toxic in high concentrations to Nitrosomonas (33).

The next lower stratum of the column is probably devoted to the conversion of ammonium ion to nitrite ion by Nitrosomonas, with the concomitant lowering of the pH value; provided, of course, that this action is not inhibited by toxic concentrations of undissociated ammonium hydroxide or by pH values that are too high for Nitrosomonas bacteria.

Finally, it is to be expected that Nitrobacter organisms would predominate in the lowest stratum of the filter, if it is deep enough, if the Nitrosomonas have not lowered the pH of the percolate to levels that are inhibitory to Nitrobacter, and if there is still some oxygen left in the interstitial atmosphere.

Granted, the demarcation between these three zones of reaction may not be very sharp and some of the Nitrobacter may coexist symbiotically with the Nitrosomas, which in turn may occur in the upper layers of hydrolysis. Nevertheless, with the hydraulic transport of nutrients under the influence of gravity, it is reasonable to expect some degree of stratification. Furthermore, it is logical to anticipate that some filters may not be deep enough, or the hydraulic loading may be too great, to enable Nitrobacter to function effectively.

To achieve more even distribution of the Nitrosomonas and Nitrobacter through the filter, and to minimize stratification with respect

to pH values, it would be well to experiment further with recirculation of part of the effluent percolate to the head of a column. A few attempts were made at repeated recycling of concentrated urine (see Section 4.07) without success, but future investigators may wish to explore this avenue more thoroughly.

6.05 Granulated Activated Carbon Columns.

The surface area for adsorption and retention of ammonium ions is much greater for activated carbons than for silica sand of comparable grain size. It is reasonable to believe, therefore, that more ammonium ions could be accommodated by adsorption and held longer for nitrification in activated carbon than in the same volume of silica sand. The results of tests with 10-percent and 5-percent natural urine percolated through activated carbon are presented in Tables 6.9 and 6.10 respectively.

By comparison of these results with those for silica sand, it is readily apparent that nitrification is more complete in the activated carbon columns as measured by the lower residual ammonia nitrogen in the effluent, the lower pH values, and the almost complete conversion of nitrites to nitrates. As might be expected, the pH values in the percolate were lower with the stronger feed, but the percentage of residual ammonia was not much higher than with the weaker feed. Apparently the adsorptive capacity and retention time were more critical factors than the inhibiting action of high ammonium ion concentrations, at least for these two dilutions. In the test of 12 March 1968 with the 10-percent urine, it is probable

Table 6.9

NITRIFICATION OF NATURAL URINE AT 10:1 DILUTION
COLUMN NO. 1---ACTIVATED CARBON

Physical data.

Diameter of column	1.25	inches
Depth of media	60	inches
Media:----activated carbon		
Grain size approx.	0.56	mm
Hydraulic loading rate	200	ml per day = 25.2 cm per day
Dosing frequency	4	times per day
Quantity per dose	50	ml
Air flow rate	7400	ml per day

Strength of natural urine at 10:1 dilution.

Kjeldahl nitrogen	760	mg N per liter
pH value	5.8	

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	3.8	3.4	4.2	3.8
Ammonia, mg N per liter	300	225	230	252
Nitrite, " " " "	0	10	0	3
Nitrate, " " " "	410	470	380	420
Total N, " " " "	710	705	610	675
Unaccounted for N, %	6.6	7.2	19.8	11.2
Ammonia, % of effluent N	42.3	31.9	37.7	37.3
Effluent ammonia, % of input N	39.5	29.6	30.2	33.2

Table 6.10

NITRIFICATION OF NATURAL URINE AT 20:1 DILUTION
COLUMN NO. 2---ACTIVATED CARBON

Physical data.

Diameter of column	1.25	inches
Depth of media	60	inches
Media: ---activated carbon		
Grain size approx.	0.56	mm
Hydraulic loading rate	200	ml per day = 25.2 cm per day
Dosing frequency	4	times per day
Quantity per dose	50	ml
Air flow rate	7400	ml per day

Strength of natural urine at 10:1 dilution.

Kjeldahl nitrogen	380	mg N per liter
pH value	5.8	

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	5.0	6.4	3.7	5.1
Ammonia, mg N per liter	140	140	100	127
Nitrite, " " " "	0	0	0	0
Nitrate, " " " "	230	215	265	237
Total N, " " " "	370	355	365	363
Unaccounted for N, %	2.6	6.6	4.0	4.5
Ammonia, % of effluent N	37.8	39.4	27.4	35.0
Effluent ammonia, % of input N	36.8	36.8	26.3	33.4

that the air supply was inadequate and that some denitrification was occurring.

Although these tests show granulated activated carbon to be more effective than sand in nitrification of 10-percent urine, it is impractical to consider activated carbon in lieu of sand for biochemical stabilization of raw urine on spacecraft. When the adsorptive capacity of the activated carbon is reached, such columns cannot be expected to perform much better than sand filters. If activated carbon is to be used for reclamation of urine, it is better that it be conserved for polishing action to remove tastes and odors from the condensate of evaporative processes.

6.06 Calcium Carbonate Column.

As shown in Table 6.11, the grain-size distribution in the calcium carbonate column was quite broad. This medium was tried in order to assess its buffering action to prevent wide fluctuations in pH arising from hydrolysis and from nitrification.

With respect to the percentage of residual ammonia nitrogen in the effluent, the calcium carbonate column performed mid-way between the silica sand and activated carbon filters. Similarly, the conversion of nitrite to nitrate was better than in sand, but not as complete as for activated carbon. The percentage of unaccounted-for nitrogen was especially high, leading one to suspect that denitrification was occurring, especially since loss of ammonia gas should be low at the pH values observed here. With the wide distribution of grain sizes, it is probable that many of the interstices were blocked to effective transfer of oxygen in air.

Table 6.11

NITRIFICATION OF NATURAL URINE AT 10:1 DILUTION
COLUMN NO. 3---CALCIUM CARBONATE

Physical data.

Diameter of column	1.25	inches
Depth of media	60	inches
Media:---calcium carbonate		
Grain size max.	10.0	mm
Grain size min.	0.2	mm
Hydraulic loading rate	80	ml per day = 10.1 cm per day
Dosing frequency	4	times per day
Quantity per dose	20	ml
Air flow rate	7400	ml per day

Strength of natural urine at 10:1 dilution.

Kjeldahl nitrogen	760	mg N per liter
pH value	5.8	

Effluent analyses.

Determination	Date 2/16/68	2/23/68	3/12/68	Average
pH value	6.3	6.3	6.5	6.4
Ammonia, mg N per liter	185	290	185	220
Nitrite, " " " "	0	0	265	88
Nitrate, " " " "	240	215	230	228
Total N, " " " "	425	505	680	536
Unaccounted for N, %	44.0	33.6	10.5	29.5
Ammonia, % of effluent N	43.5	57.5	27.2	41.8
Effluent ammonia, % of input N	24.4	38.2	24.4	29.0

6.07 Removal and Stabilization of Carbonaceous Matter (COD).

The removal of chemical oxygen demand from natural urine in ten columns is shown in Table 6.12. With the granulated activated carbon media, as might be anticipated from its high adsorptive capacity, the removal of COD in the six tests ranged from 91.5 to 100 percent, with a 6-test average of 96 percent.

For the silica sand columns that received 10:1 dilutions of natural urine, the removal of COD ranged from 71 to 85 percent with a 12-test average of 79 percent. Removal at the stronger 5:1 dilution was slightly better and from the weaker 20:1 dilution slightly poorer than for the 10:1 dilution. For the total of 18 tests with silica sand, the average removal was 79 percent. The same efficiency was achieved with the calcium carbonate media. These values compare quite favorably with the 86 percent removal of COD from municipal sewage (see Table 6.1).

The results with synthetic urine are anomalous. Theoretically the COD of the synthetic urine used in this study, containing no glucose and only 8600 mg per liter of urea nitrogen in addition to the mineral salts, should have been zero; yet the percolate contained an average of 85 mg per liter of COD. This paradox can be explained by three effects, all of which probably occurred simultaneously. First, the zero COD of the synthetic urine was theoretical but not necessarily operational because the high chloride content may have produced some interference and apparent COD despite the use of mercuric sulfate, as explained in Standard Methods (34). With a synthetic mineralized seawater containing no organic matter, it is

Table 6.12

REMOVAL OF CHEMICAL OXYGEN DEMAND
FROM NATURAL AND SYNTHETIC URINE

Column No.	Percent Dilution	Packing Medium	Effluent COD, mg/liter 2/16/68	2/23/68	3/12/67	Average	Percent Removed*
1	10	Carbon	22	15	0	12	98
2	5	Carbon	22	32	15	23	94
3	10	$CaCO_3$	150	220	110	160	79
4	10	Sand	185	145	110	147	80
5	10	Sand	200	130	110	147	80
6	10	Sand	85	90	80	85	--
7	10	Sand	115	145	145	135	82
8	10	Sand	215	220	140	192	74
9	20	Sand	255	290	290	278	81
10	5	Sand	100	90	85	92	76

*Based on a COD of 7500 mg per liter for undiluted natural urine. The theoretical COD of the synthetic urine used in these tests (no glucose) was zero.

not uncommon to observe apparent COD values of 30-35 mg per liter. Second, the columns had been ripened for four months with settled municipal sewage, from which some previously adsorbed COD may have leached into the percolate. Third, the Nitrosomonas and Nitrobacter are autotrophic bacteria that synthesize photoplasm from ammonia and bicarbonates. Some of these bacterial bodies undoubtedly appeared in the percolate and exerted COD.

6.08 Odor and Color.

The odor of the percolate from the silica sand and calcium carbonate columns was not unpleasant. It resembled the odor from a well-oxidized biologically treated municipal sewage. No foul odors characteristic of an anaerobic condition were detected, even at the 20-percent dilution. The effluents from the activated carbon columns were completely free from any odor.

The percolates from the sand and calcium carbonate columns were free from visual turbidity but retained a yellow color comparable to that of the dilution applied. From the activated carbon columns, the effluents were crystal clear and devoid of color.

6.09 Field Tests for Nitrates.

In the southern part of Los Angeles County there are many acres of porous soil that have been used as cattle feed pens for a decade or longer. Except during rainy weather, the only liquid reaching and percolating through the soil at these feed pens is the urine from cattle. It was interesting to determine, therefore, whether or not a signficant degree of nitrification was occurring in the soil moisture

at these locations, where the opportunities for seeding and ripening were more than adequate.

Core samples were taken near the feed troughs where the density of cattle was greatest and micturition most frequent. These core samples, from depths of 6 inches to 48 inches, were placed in plastic bags to minimize loss of moisture, transported to the laboratory, and analyzed immediately for soil moisture and nitrate nitrogen. The results, presented in Table 6.13, show that the

Table 6.13
NITRATE NITROGEN IN SOIL AT CATTLE FEED PENS

	Soil Depth, inches	Soil Moisture, Percent	Nitrate N, mg per liter of soil moisture
Sample I	12	10.0	33
	24	5.5	72
	36	5.2	72
	48	5.0	142
Sample II	5	49.8	68
	12	9.6	68
	21	8.0	160
	30	3.5	115
	42	2.0	60

moisture content of the soil was quite low, except near the surface, which leads to the conclusion that drainage beyond a foot of depth was rapid and that the soil had a very low specific retention of pellicular and interstitial water. The table also indicates that the

nitrate nitrogen concentrations in the soil moisture were also minimal in relation to the probable nitrogen content of bovine urine (which was not assayed, but presumably was comparable to that of human urine, i.e. about 10,000 mg per liter of nitrogen). Since there had been no recent rains during this period of the year, it is apparent that only an insignificant portion of the organic and ammonia nitrogen had been oxidized to nitrate nitrogen. Indeed, some of this oxidation may have occurred during the coring and transporting operations, when the core was exposed to atmospheric oxygen.

CHAPTER VII
SUMMARY

It was the objective of this investigation to study the oxidation of nitrogenous and carbonaceous organic matter in urine by intermittent percolation through porous media. From previous research at the California Institute of Technology and from decades of field experience by others, it was known that much of the organic matter in municipal sewage could be converted to nitrates, sulfates, bicarbonates, and other oxidized stable compounds by properly controlled intermittent filtration through sand; but to the best of our knowledge no one had ever studied similar stabilization of full-strength or partially diluted urine. If a high degree of oxidation could be accomplished, this phenomenon might find applications in the reclamation of urine in space vehicles or at lunar stations.

The results of the experiments described in this report are, for the most part, largely discouraging insofar as the primary objective is concerned. At concentrations stronger than 20-percent urine in distilled water, nitrification by percolation through sand was virtually nil. At weaker dilutions (5 and 10 percent) some nitrification was evident but much of the nitrogen came through the columns as nitrite rather than as nitrate. About 80 percent of the carbonaceous material, as measured by the COD test, was removed by adsorption and subsequently oxidized by heterotrophic bacteria. Clogging of the upper two or three inches of sand by chemical precipitates and biological growths proved to be a major handicap in that it militated against proper reaeration of the sand interstices,

contrary to the maintenance of an aerobic environment so necessary for effective nitrification.

Higher degrees of nitrification and better removal of COD from 5- and 10-percent urine solutions were achieved when granular activated carbon was used in the columns, but such media would be impractical for space travel unless facilities for reactivation were available. Calcium carbonate particles were also tried as media, but proved to be not much more effective than silica sand.

The project did provide considerable insight into the mechanisms of biochemical stabilization of urine. On the basis of these experiments and within the limitations of the systems tested, the following observations are advanced:

1. The hydrolysis of urea and the ammonification of most other nitrogenous compounds in natural urine were substantially complete after percolation through 24 inches of medium silica sand.

2. Hydrolysis and ammonification raised the pH value of the percolate from about 5.8 to about 8.8 or 9.0.

3. At pH 9, about 35 percent of the ammonia nitrogen is in the form of undissociated ammonium hydroxide, which is considered to be highly toxic to many bacteria. Consequently much of the inhibition to conversion of dissociated ammonium ions to nitrates may be attributed to the toxic action of high concentrations of undissociated ammonium hydroxide.

4. Ammonium ions are readily adsorbed by the abundant surface areas in fine porous media and held in the pellicular and interstitial water until they are oxidized to nitrite and nitrate ions, which are then

quickly leached by percolating water. Nitrification, however, is not a rapid process even under ideal conditions; hence sufficient time in the adsorbed state must be available to enable ammonium ions to be oxidized to nitrites or nitrates.

5. Adsorption and retention are much better in granular activated carbon than in silica sand or calcium carbonate media.

6. If the adsorptive capacity of the media is great enough, if sufficient time in the adsorbed state is available, if the daily volume of percolate does not exceed the total volume of pellicular and interstitial water, if an aerobic environment is maintained, and if the concentration of undissociated ammonium hydroxide is not unduly toxic, a high degree of nitrification can be achieved by intermittent filtration through porous media. It would be difficult, however, to satisfy these multiple conditions in a practical unit for space travel.

7. The maintenance of an aerobic environment in the interstices of the media is absolutely essential for complete nitrification. The diffusion of atmospheric oxygen into and through a sand column is diminished and sometimes stopped by clogging of the surface and interstices by biological growths and chemical precipitates. For the concentrations of urine used in these tests, a forced draft of air was necessary, but not always sufficient, to maintain aerobiosis.

8. When Nitrosomonas bacteria succeed in converting ammonium ions to nitrites, the released hydrogen ions may lower the pH of the percolate sufficiently to inhibit Nitrobacter in lower strata of the filter from converting nitrites to nitrates. This inhibition may be aggravated by insufficient oxygen, especially in clogged interstices.

9. Even when nitrification to nitrites or nitrates is achieved, there is a possibility that denitrification to nitrogen gas may occur if the lower strata of the filter become anaerobic. Such production of nitrogen may be beneficial in space vehicles, to replace atmospheric nitrogen that is lost from a vehicle into the surrounding vacuum.

10. With silica sand or calcium carbonate media, about 80 percent of the carbonaceous material in diluted urine can be removed by adsorption and subsequently oxidized. With granular activated carbon, the efficiency of removal is even higher.

APPENDIX A

REFERENCES CITED

1. Webb, Paul (Editor), "Bioastronautics Data Book," STID, NASA (1964).

2. Breeze, R. K., "Space Vehicle Environmental Control Requirements Based on Equipment and Physiological Criteria," ASD Tech. Rept. 61-161 Part I, Wright-Patterson AFB, (Dec. 1961).

3. Wallman, H. and Barnett, S. M., "Water Recovery Systems (Multi-Variable), WADD Tech. Report 60-243 (March 1960).

4. Armstrong, R. C., "Life Support System for Space Flights of Extended Time Periods," NASA CR-614 (Nov. 1966).

5. Rudek, Fred P. "Water Reuse in Space," AIChE Symposium Series No. 78 (Water Reuse), Vol. 63, p. 210 (1967).

6. Mechalas, B. J., Geckler, R. P., Ducey, F. D., and Farquhar, R. D., "Water and Waste Management in Spacecraft," Aerojet-General Corporation, presented at 38th Annual Meeting of Calif. Water Poln. Control Assn. (April 1966).

7. Rich, L. G., Ingram, W. M., and Berger, B. B., "Waste Disposal on Space Craft and Its Bearing on Terrestrial Problems," USPHS Publication No. 999-WP-29 (Aug. 1965).

8. McKee, J. E., "Liquid Wastes and Water Potability in Space Vehicles," Conf. on Nutrition in Space and Related Waste Problems, NASA SP-70, p. 273, Tampa, Fla. (April 1964).

9. Altman, P. L., "Blood and Other Body Fluids," Ch. XIX and Urine Table 122, Comm. on Biological Handbooks, Fed. Am. Soc. Exp. Biol., Wash., D.C. (1961).

10. Wallman, H. and Steele, J. A., "Water Reclamation Subsystems for a Space Station," AIChE Symposium on Aerospace Life Support, 53rd National Meeting, Pgh., Pa. (May 1964).

11. Cushing, A. R., "The Secretion of the Urine," Longman, Green and Co., Ltd., London (1956).

12. Spector, Wm. S. (Ed.), "Handbook of Biological Data," p. 341, W. B. Saunders Co., Phila. (1956).

13. Allen, A. C., "The Kidney, Medical and Surgical Diseases," p. 87, 2nd Ed., Grune and Stratton, N.Y. (1962).

14. Collins, V. G. and Popma, D.C., "Water Reclamation and Conservation in a Closed Ecological System," L-4971, NASA Langley Research Center, presented at the Ecological Technology Symposium, Wash., D.C. (Feb. 1966).

15. Ingram, W. T. "Survival in a Capsule," Chemistry in Canada, p. 86 (Nov. 1959).

16. Morris, J. C. (Chairman), "Water Quality Standards for Long Duration Manned Space Missions," Report of an Ad Hoc Panel, Space Science Board, NAS/NRC, (Sept. 1967).

17. Anon., "Public Health Service Drinking Water Standards," USPHS Publication No. 956, Washington, D.C. (1962).

18. Anon., "International Standards for Drinking Water," World Health Organization, Geneva, Switzerland (1958).

19. Anon., "European Standards for Drinking Water," World Health Organization, Geneva, Switzerland (1961).

20. Johnson, Robert E., "Human Nutritional Requirements for Water in Long Space Flights," Conf. on Nutrition in Space and Related Waste Problems, NASA SP-70, p. 159, Tampa, Fla. (April 1964).

21. Wallman, H., Steele, J. A., and Lubitz, J. A., "Multi-Filter System for Water Reclamation," 35th Aerospace Medical Assoc. Meeting, Miami, Fla. (May 1964).

22. Ingram, W. T., "Microbiological Waste Treatment Processes in a Closed Environment," Tech. Doc. Report No. AMRL-TDR-62-126, USAF (Nov. 1962).

23. McElroy, Wm. D. and Glass, Bentley (Ed.), "Inorganic Nitrogen Metabolism," 728 pp., The Johns Hopkins Press, Baltimore (1956).

24. Quastel, J. H. and Scholefield, P. G., "Biochemistry of Nitrification in Soil," Bacteriological Reviews, 15, 1 (1951).

25. Preul, H. C. and Schroepfer, G. J., "Travel of Nitrogen in Soils," Jour. Water Pollution Control Fed., 40, 30 (1968).

26. Bailey, Geo. W., "Role of Soils and Sediment in Water Pollution Control, Part I. Reactions of Nitrogenous and Phosphatic Compounds With Soils and Geologic Strata," Southeast Water Laboratory, FWPCA, U. S. Dept. of Interior (March 1968).

27. Conrad, J. P., "Hydrolysis of Urea by Thermolabile Catalysis," Soil Sci., 49, 253 (1940).

28. Conrad, J. P., "The Nature of the Catalyst Causing the Hydrolysis of Urea in Soils," Soil Sci., 50, 119 (1940).

29. Stout, P. R., Burau, R. G., and Allardice, W. R., "A Study of the Vertical Movement of Nitrogenous Matter from the Ground Surface to the Water Table in the Vicinity of Grover City and Arroyo Grande--San Luis Obispo County," Central Coastal Regional Water Pollution Control Board, San Luis Obispo, Calif. (Jan. 1965).

30. Jensen, H. L., "Effect of Organic Compounds on Nitrosomonas," Nature, 165, 974 (1950).

31. Lees, H., "Hydroxylamine As an Intermediate in Nitrification," Nature, 169, 156 (1952).

32. Hofman, T. and Lees, H., "The Respiration and Intermediate Metabolism of Nitrosomonas," Biochem. Jour. 54:4, 579 (1953).

33. Meyerhof, O., "Untersuchungen über den Atmungsvorgang nitrifizierenden Bakterien," Pflüger's Archiv für gesamte Physiologie, 164, 353 (1916); 165, 229 (1916); and 166, 240 (1917).

34. Anon., "Standard Methods for the Examination of Water and Wastewater," 12th Edition, 769 pp., Am. Public Health Assn., New York, (1965).

APPENDIX B

OXYGEN RELATIONSHIPS IN INTERMITTENT SAND FILTRATION OF WASTEWATERS

Thesis by

Albert Bernard Pincince

In Partial Fulfillment of the Requirements

For the Degree of

Doctor of Philosophy

California Institute of Technology

Pasadena, California

1968

(Submitted July 24, 1967)

ACKNOWLEDGMENTS

Throughout graduate study at Caltech and particularly during the conduct of research and preparation of this thesis, I was assisted and encouraged by many people.

As my advisor for almost four years, Dr. Jack E. McKee guided me in the selection of courses and directed me through my research.

Dr. James J. Morgan made numerous helpful suggestions during the research and reviewed the first five chapters of this thesis.

Studies with the Warburg apparatus and some of the field studies were conducted with Mr. Dennis R. Kasper.

Three subroutines used in the computer programs were written by Dr. Peter Rüst, who patiently explained their use and the proper calling sequences.

Test apparatus was designed and constructed with the assistance of Mr. Elton F. Daly and Mr. Robert L. Greenway.

Mr. Carl Green drew most of the figures.

Drafts of this report were ably typed by my wife Barbara and Mrs. Peggy Freeland. The final copy was typed by my wife.

The cooperation and assistance of all these people is deeply appreciated.

This research was supported primarily by a Public Health Service Training Grant and partially by the National Aeronautics and Space Administration (Research Grant NCR-05-002-036). The generous help of these sponsors is gratefully acknowledged.

Finally, my wife and our two children, Tommy and Cathy, deserve special mention for bearing the impositions of a student's life.

ABP

ABSTRACT

A model for some of the many physical-chemical and biological processes in intermittent sand filtration of wastewaters is described and an expression for oxygen transfer is formulated.

The model assumes that aerobic bacterial activity within the sand or soil matrix is limited, mostly by oxygen deficiency, while the surface is ponded with wastewater. Atmospheric oxygen reenters into the soil after infiltration ends. Aerobic activity is resumed, but the extent of penetration of oxygen is limited and some depths may be always anaerobic. These assumptions lead to the conclusion that the percolate shows large variations with respect to the concentration of certain contaminants, with some portions showing little change in a specific contaminant. Analyses of soil moisture in field studies and of effluent from laboratory sand columns substantiated the model.

The oxygen content of the system at sufficiently long times after addition of wastes can be described by a quasi steady-state diffusion equation including a term for an oxygen sink. Measurements of oxygen content during laboratory and field studies show that the oxygen profile changes only slightly up to two days after the quasi-steady state is attained.

Results of these hypotheses and experimental verification can be applied in the operation of existing facilities and in the interpretation of data from pilot plant-studies.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

CHAPTER 1

INTRODUCTION

1-1 Background

Intermittent sand filtration is a method of wastewater treatment in which the liquid is applied intermittently to a natural or artificial bed of porous media and allowed to percolate through it to underdrains or to the ground-water table. Its use for the disposal of wastewaters is by no means new. Sedgwick (1) and Kinnicutt, Winslow, and Pratt (2) refer to laboratory studies by Sir Edward Frankland published in 1870 by the Rivers Pollution Commission of Great Britain. These studies, the first on intermittent percolation, showed that the process produced an effluent that was "remarkably purified."

Several important conclusions were drawn from Frankland's studies. He observed that the word "filtration" was really a misnomer since not only physical but also biological processes are involved. The experimenters recognized that aerobic bacterial action was needed to oxidize wastes and to minimize clogging. Hence sewage, which is usually low in dissolved oxygen, had to be applied intermittently to allow atmospheric oxygen to reach the microoganisms.

The first practical application of intermittent sand filtration was by J. Bailey-Denton who constructed four

beds totalling 20 acres at Merthyd Tydvil, Wales in 1871. Raw sewage was applied to each bed for six hours daily at a net rate of 60,000 gallons per acre per day (0.18 ft per day). The effluent was collected by an underdrain system. Application was later reduced to 16,000 gallons per acre per day by addition of more land. The original plant worked well, however, and it was a practical demonstration of Frankland's process (2).

The first studies in the United States on intermittent sand filtration were conducted at the Lawrence (Massachusetts) Experiment Station, beginning in 1887. Results indicated again the need for intermittent application of sewage and showed a significant reduction in bacterial numbers. As a result of these studies, the towns of Framingham and Brockton soon built large municipal filters and other cities followed suit(1).

The many studies on intermittent sand filtration since Frankland's original experiments have well established the need for intermittent application of wastes to allow sufficient atmospheric aeration and to avoid clogging. Spreading basins operated continuously become anaerobic because any dissolved oxygen that may be present in wastewater is insufficient to satisfy the biochemical oxygen demand.

Although the importance of sufficient aeration during waste degradation by intermittent percolation has

been accepted by sanitary engineers, the phenomena associated with reoxygenation remain obscure. With the increased interest in tertiary wastewater treatment and in the recharge of ground-water basins for water reclamation, the mechanisms and rates by which oxygen is utilized during percolation and resupplied from the surface deserve attention.

Adequate descriptions of the physical processes are equally lacking. Most published studies approach intermittent sand filtration as a "black box," recording only changes in effluent quality caused by variations in influent type, loading and strength, and frequency of application. There have been few attempts to interrelate physical and biological processes, but such a description is necessary to understand transfer and bacterial utilization of oxygen.

It was the aim of this research, therefore,

1. To investigate deoxygenation and reaeration in soil systems used for stabilization of wastewaters, and
2. To describe other processes in intermittent sand filtration as a prelude for understanding oxygen transfer and bacterial utilization.

1-2 Use of Intermittent Sand Filtration

Early intermittent sand filters treated either raw or settled sewage or septic-tank effluent. Since these filters required large land areas (e.g. about five acres per 1000 people) they were largely replaced, as populations increased, by trickling filters or other processes requiring less land. In recent years, the need for ground-water replenishment and for advanced waste treatment has rekindled interest in intermittent sand filtration. When used for these purposes, intermittent sand filtration follows secondary treatment. Hence, the future of this process appears to lie in tertiary, rather than primary treatment.

Intermittent sand filtration following secondary treatment is being used successfully for ground-water recharge in several locations in the United States and other countries. Noteworthy examples are installations at Whittier Narrows, California and Tel Aviv, Israel.

The basins at Whittier Narrows are part of a demonstration project for reclaiming domestic wastewaters by activated-sludge treatment followed by surface spreading for ground-water recharge. Approximately 15 million gallons per day (mgd) of domestic wastes are presently being treated.

The Israel project is designed to reclaim wastewater from the Tel Aviv metropolitan area. Wastes are treated in stabilization lagoons before ground-water recharge through dune sands (3). The facility is being planned for the year 1990 when the design population will exceed one million people and expected flows will be over 100 million cubic meters per year (72 mgd).

Advanced waste-treatment methods are needed to remove nutrients such as nitrogen and phosphorus from wastewaters in order to avoid algal blooms in receiving streams. Intermittent sand filtration may be used as the first step in a nitrification-denitrification scheme for removing nitrogen from wastewaters because it encourages the growth of nitrifying bacteria. Because intermittent sand filtration does not remove phosphorus, some other operation will be needed for phosphorus removal. Levin (4) has described modifications to the activated-sludge process for phosphorus removal.

1-3 Clogging

Since infiltration rates tend to decrease as a result of clogging, consideration of the causes of clogging is required to understand the necessity for intermittent application. Allison (5) showed that clogging was not due to purely physical causes such as soil-aggregate breakdown but was caused by microorganisms. Gupta and Swartzendruber (6) confirmed Allison's work when they found that hydraulic

conductivity for boiled deionized water decreased markedly during flow and that no decrease occurred when a 0.1 percent phenol solution was used. In addition, no clogging took place when bacteria at the soil surface were less than 400,000 per gram of soil, but that drastic reduction occurred above this concentration. Since the volume of bacteria constitutes only a small portion of the pore space, Gupta and Swartzendruber (6) concluded that associated products rather than the bacteria themselves cause clogging.

McGauhey and Winneberger (7), studying septic tank percolation systems, claimed that sulfide formation in soil systems caused clogging through precipitation of ferrous sulfide. Nevo, with Avnimelech (8) and with Mitchell (9), passed NH_4NO_3 in tap water through sands to which organics had been added. They disputed McGauhey and Winneberger's claim and believed that sulfide formation was only an indicator of anaerobic conditions. Mitchell and Nevo (9) stated that polysaccharide accumulation was highly correlated to clogging, but Avnimelech and Nevo (8) concluded that polyuronides were responsible.

Thomas, Schwartz, and Bendixen (10) found that phosphate and total organic matter (chemical oxygen demand) accumulated in soil during clogging, in addition

to iron, sulfide, polysaccharide, and polyuronide. Of these parameters, only organic matter declined as the infiltration rate was partially recovered during a rest cycle.

The substances causing clogging are most readily degraded aerobically by fungi and bacteria. Because the oxygen demand of wastewaters is many times the dissolved oxygen (if any) in the wastewater, oxygen must be supplied to the soil system. The most practical way to do so is to allow air to enter the system during a rest cycle.

Amramy (3) notes that while many of the substances causing clogging are produced anaerobically, polysaccharides cannot be produced without oxygen. Since polysaccharides accumulate even during aerobic conditions, intermittent operation of percolating beds serves still another function, namely, to force the bacteria to respire endogenously and thereby to utilize and stabilize some of their own waste products.

1-4 Spreading Basin Operation

In practice, effluent from a primary or secondary wastewater treatment plant is spread on a basin and the basin is allowed to drain until no liquid is ponded on the surface. The basin is then "rested" before further application of treated wastes to allow oxygen from the atmosphere to diffuse into the soil.

The cycle time (ponded plus resting) for a given installation has been determined in the past by trial and error from observations of clogging rates and effluent quality. These studies have resulted in a variety of loading methods.

Amramy (3), applying effluent from a series of lagoons on a dune sand (effective size 1.15 mm), could maintain an approximately constant infiltration rate by using a cycle of ten days, five wet and five resting. For safety, he suggested five or six wet days and twice as many resting days.

Robeck et al (11) noticed that effluent quality was improved by reducing the total cycle time to between four and eight hours.

Others, e.g. Laverty, Stone, and Meyerson (12) and McMichael and McKee (13) used 24-hour cycles with success. McMichael and McKee maintained the infiltration rate in test basins by adjusting the volume spread once each day so that the wet time did not exceed 12 hours.

These examples indicate that installations have successfully used cycles of between four hours and about two weeks. The results of this research should provide some insight into the required cycle times.

CHAPTER 2

PROCESSES IN INTERMITTENT SAND FILTRATION

2-1 Redistribution of Soil Water

If Darcy's law is assumed valid for unsaturated flow of an incompressible liquid in porous media, continuity may be written for vertical flow as

$$\frac{\partial \theta}{\partial t} = \frac{\partial}{\partial z}\left(K \frac{\partial p}{\partial z}\right) - \frac{\partial K}{\partial z} \qquad (2\text{-}1)$$

where

θ = soil water content, L^3/L^3*

t = time

z = distance measured positively downward, L

K = capillary conductivity, L/t

p = soil water pressure expressed as height of water, L

Equation 2-1 is difficult to solve because both K and p (sometimes called capillary head) depend on moisture content. The problem is compounded because the relation between θ and p is not single-valued but depends on the history of the media (14). Equation 2-1 has been solved

---

* Dimensions are given in terms of mass (M), length (L), and time (t).

only for the simplest cases. Even then, solutions have required simplifying assumptions or the use of digital computers.

Some work has been done on the problem of water infiltrating a soil (15, 16) and on drainage of initially saturated porous materials (17, 18, 19), but the literature on redistribution of soil water after infiltration is scarce indeed. Biswas, Nielsen, and Biggar (14) have observed that the distribution pattern depends on soil type and depth of water added (Fig. 2-1). For coarse materials or large additions of water, the medium was saturated to a short depth as infiltration ended. Further redistribution proceeded with an immediate moisture loss near the surface and a reduction in the zone of saturation (Fig. 2-1a). For fine-textured soils whose retained moisture varied gradually with increasing height above a water table or for shallow depths of water, moisture contents never approached saturation, except at the soil surface (Fig. 2-1b). For fine-textured soils, infiltration (the passage of water into the soil across the interface of water and soil, or air and soil) is often the limiting or controlling mechanism rather than percolation (the vertical downward movement of water within the soil).

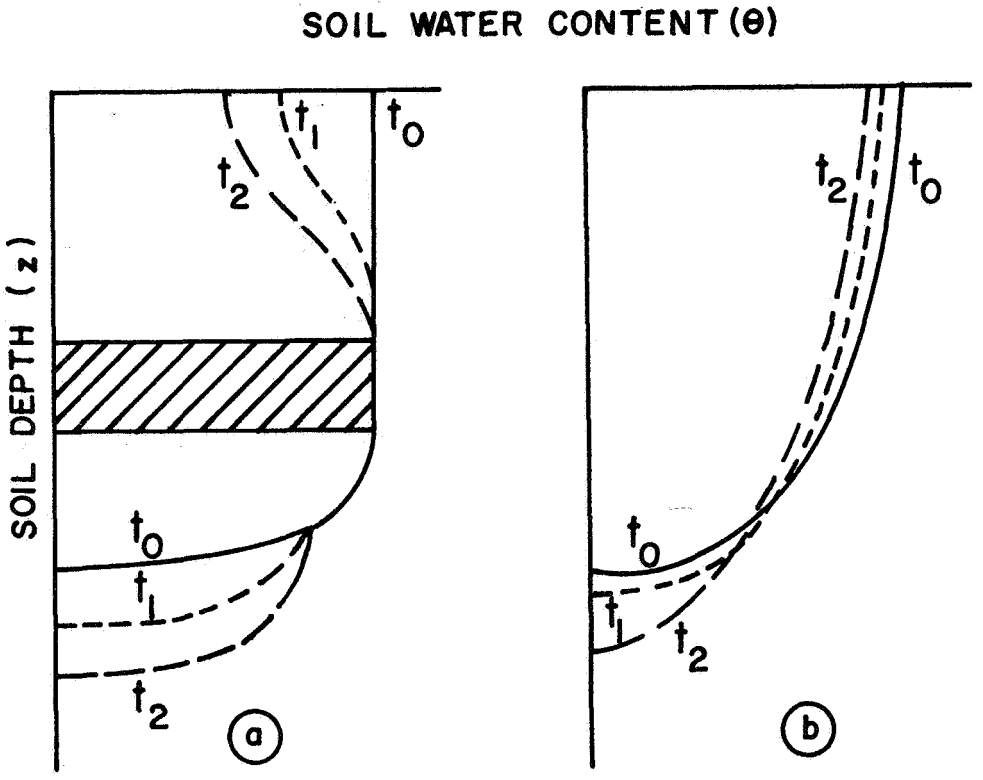


Fig. 2-1 Hypothetical soil-water profiles during redistribution of water. In a, the sand is coarse; in b, the sand is fine. At time t_0, for profile a, the soil is saturated from the surface to the lower edge of the cross-hatched area. At time t_2, the zone of constant water has diminished to that represented by the cross-hatched area. After Biswas, Nielsen, and Biggar (14).

2-2 Mechanisms for Removal of Pollutants

Processes acting on pollutants during intermittent sand filtration may be divided into two categories, viz. physical-chemical processes that remove pollutant from the percolate and biological processes that stabilize the wastes. These two categories are reviewed briefly in the following sections.

2-2-1 Removal of Wastes from Percolating Liquid

In the early days of intermittent sand filtration when raw sewage or settled sewage was applied to sand beds, straining was an important removal mechanism and the suspended matter thus retained tended to form a mat on the filter bed. This build-up of organic matter impeded infiltration and hindered oxygen transfer into the soil. Now, when intermittent sand filtration is used it generally follows secondary treatment and constitutes advanced waste treatment or initiates wastewater reclamation processes for replenishment of the ground water. Suspended-solids content of the typical water presently used for wastewater reclamation is substantially below that of raw sewage or primary effluent. Hence, suspended-matter removal by intermittent sand filtration is less important now than in former years.

While the soil is draining, some polluting material is removed from the percolating liquid by physical adsorption on soil surfaces, some by diffusion into stagnant zones, and some by biological assimilation and biosynthesis.

Concurrently, other substances are being desorbed from soil surfaces, removed from stagnant zones, and excreted as a result of biological metabolism. In intermittent sand filtration the substances being adsorbed or otherwise removed from percolating water may be dissolved, colloidal, or suspended solids. In general, they are complex organic compounds or ammonium ions. The substances being returned to the percolating water are generally dissolved minerals or stabilized organics.

The extent of biological utilization of organic substances during percolation depends both on the nature of the component being considered and on oxygen availability. Substances that are readily biodegradable will be almost completely stabilized while oxygen is still present. More resistant compounds may be adsorbed by biological films and metabolized slowly. Some compounds may be essentially unaffected by percolation through biologically rich porous media.

2-2-2 Biological Processes

The object of intermittent sand filtration, in addition to maintaining high rates of hydraulic acceptance, is to stabilize wastes to innocuous end products.

Under aerobic conditions, the major end products in biochemical stabilization of carbonaceous and nitrogenous substances are water,carbon dioxide, bicarbonates, sulfates, phosphates and nitrates. In the absence of oxygen, carbonaceous material may be converted to carbon dioxide and methane, but nitrogenous substances degrade only to ammonia, and cannot be oxidized to nitrate.

Ammonia is nitrified in two steps by autotrophic bacteria which do not use organic carbon. Nitrosomonas first mediate the oxidation of ammonia to nitrite and Nitrobacter complete the process by catalyzing the oxidation of nitrite to nitrate. Thimann (20) considers nitrite formation as a pair of linked reactions, the second of which has two alternatives:

$$2\ NH_4^+ + 4H_2O \longrightarrow 2NO_2^- + 4H^+ + 12\ (H) \qquad (2\text{-}2)$$

$$12\ (H) + 3\ O_2 \longrightarrow 6H_2O \qquad (2\text{-}3a)$$

$$12\ (H) + 3\ CO_2 \longrightarrow 3(CH_2O) + 3H_2O \qquad (2\text{-}3b)$$

The proportion of reaction 2-3a to 2-3b can be estimated from studies that measured the ratio NH_4^+ oxidized: CO_2 reduced. Thimann (20) has obtained values ranging from 20 to 100 for this ratio and cites values from 15 to 53.

A ratio of 15 corresponds to 21.5 of reaction 2-3a to one of reaction 2-3b; a ratio of 100 corresponds to 149 of reaction 2-3a to one of reaction 2-3b.

The reactions for nitrate formation can be written as:

$$2NO_2^- + 2H_2O \longrightarrow 2NO_3^- + 4(H) \qquad (2\text{-}4)$$

$$4(H) + O_2 \longrightarrow 2H_2O \qquad (2\text{-}5a)$$

$$4(H) + CO_2 \longrightarrow (CH_2O) + H_2O \qquad (2\text{-}5b)$$

Schön (21) has measured the ratio of NO_2^- oxidized: CO_2 reduced. His value corresponds to 36 of 2-5a to one of 2-5b.

In aerobic activity involving heterotrophic bacteria, oxygen is the terminal electron acceptor of the electron-transfer chain. In anaerobiosis, other elements are the acceptors and are reduced. These elements include nitrogen in nitrate and nitrite, sulfur in sulfate, and carbon in organic compounds.

Nitrate reduction (denitrification) can be either assimilatory or dissimilatory. In assimilatory denitrification, nitrogen is reduced to minus-three valence for incorporation in the cell structure. Dissimilatory nitrate reduction produces N_2, N_2O, and NO. The formation of the latter three gases would result in a reduction of total nitrogen in the soil water. The studies on wastewater reclamation by McMichael and McKee (13) did not show a

nitrogen loss although denitrification is known to occur in soils (22).

The decomposition of organic materials with sulfate as the terminal electron acceptor produces sulfide or elemental sulfur. Sulfide is formed in soil systems only after clogging has begun (10). The test basins used by McMichael and McKee (13) have not clogged although they have been in operation for more than four years. Hence it would seem that significant sulfate reduction can be eliminated in at least some intermittent filters.

Although carbon as an electron acceptor (fermentation) results in reaction rates that are considerably slower than found under aerobic conditions (23), fermentation may account for some stabilization in an intermittent sand filter because only the top few feet are aerobic and even this area becomes anaerobic after the surface is ponded.

The presence or absence of significant anaerobic bacterial activity at depths permanently devoid of oxygen should be considered when one is studying oxygen transfer during intermittent sand filtration. This matter is important because the movement of gases produced at anaerobic depths (nitrogen, methane) will influence the movement of oxygen gas into the aerobic zone. The effect of other gas movements on the movement of oxygen will be discussed below.

From measurements showing that more organic nitrogen was collected in a laboratory-filter effluent than was added, Morgan and Gilcreas (24) have suggested that nitrogen fixation occurs in the upper portion of sand filters.

2-3 A Model for the Processes in Intermittent Sand Filtration

When secondary treatment plant effluent infiltrates into a soil, substances are removed from the percolating liquid as described previously when the incoming liquid displaces whatever liquid existed previously in the soil. Although the substances in the incoming liquid are affected by bacterial activity, stabilization of some components is limited because the liquid percolates through the soil too quickly for the microoganisms to assimilate and completely degrade the wastes. In addition, bacterial activity is limited by availability of oxygen.

Therefore, immediately after ponded water has completely infiltrated into the soil, the composition of the soil water with respect to those components not assimilated or adsorbed is essentially constant for some depth into the soil. During subsequent drainage, air enters into the soil and aerobic activity increases.

Secondary effluent is complex but can be characterized by analyses for the carbonaceous and nitrogenous components (carbohydrates, fatty acids, protein). At most wastewater treatment plants, almost all of the

nitrogen is present as ammonia or organic matter; at the Whittier Narrows Water Reclamation Plant (activated-sludge plant) the effluent, which is to be used for ground-water recharge, contains 10 to 15 percent of the total nitrogen as nitrate.

The extent of aerobic activity can be related to the increase in nitrate in the soil moisture. Other chemical determinations could be used for this purpose, but nitrate determination is convenient and nitrification is an important function. Because of limited stabilization while the surface is ponded, nitrate content of soil water can be expected to be essentially constant throughout the soil profile at the end of infiltration and to increase as bacterial activity stabilizes the influent. The first increase in nitrification will occur near the surface because atmospheric oxygen is first available at the surface. Nitrification will progress toward greater depths as oxygen becomes available to nitrifying bacteria. However, there is a depth below which there is no oxygen. Incoming wastewater that passes into depths permanently devoid of oxygen does not become further nitrified.

Fig. 2-2 shows hypothesized changes in nitrate with time and depth in an intermittent filter. At t_0 infiltration has just been completed and the nitrate content of the soil water is roughly constant with depth, but some-

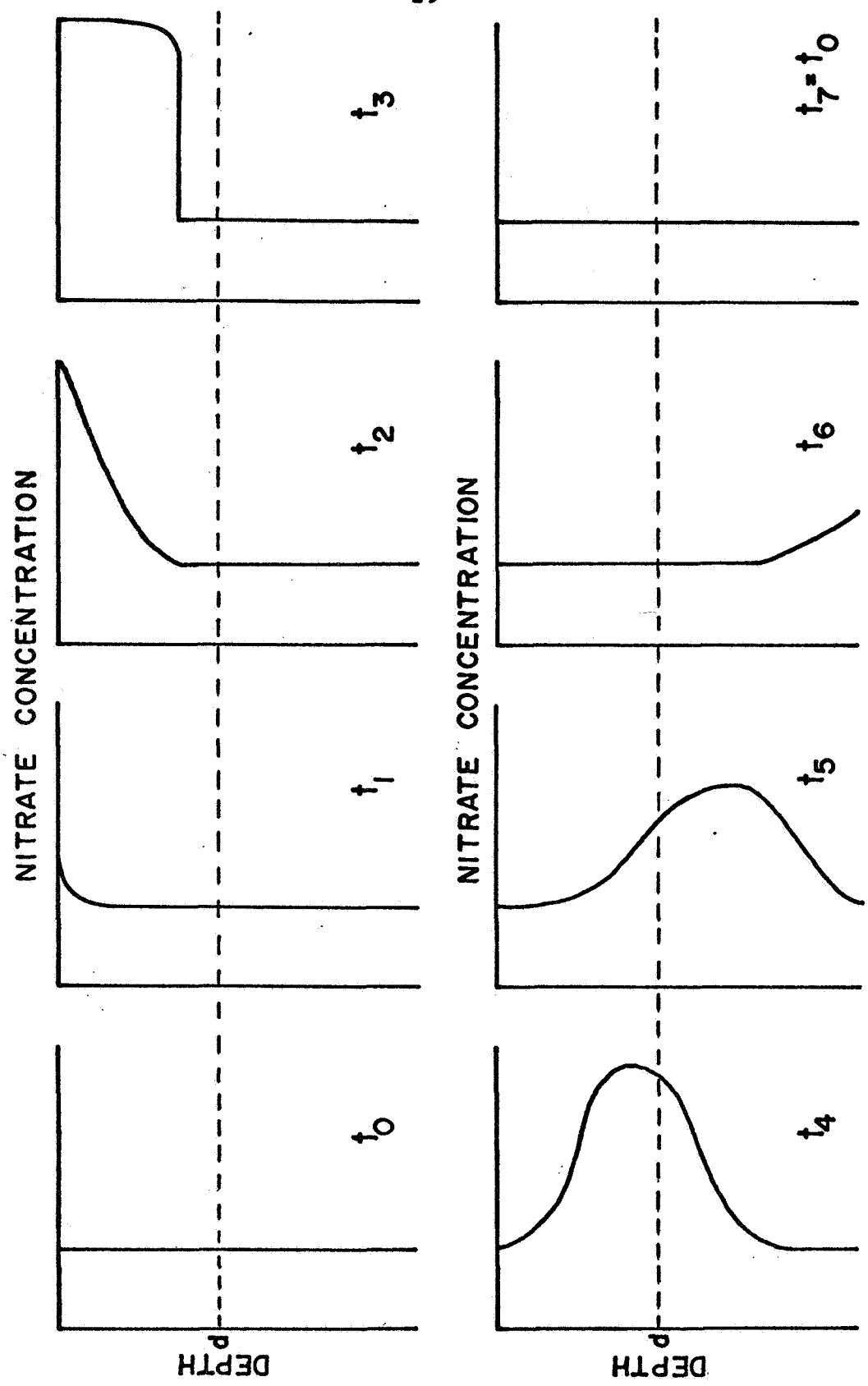


Fig. 2-2 Hypothesized variation of nitrate in soil water

what higher than the nitrate content of the secondary effluent applied because of the nitrification that occurred during drainage. The sketches for times t_1 to t_3 show the change in nitrates with time. Time t_3 is just before more wastewater is spread on the filter surface.

The incoming waste displaces the pellicular water and dispersion changes the shape of the nitrate wave, as shown in sketches for t_4 to t_7.

A plot of the variation of nitrate content at depth "d" of Fig. 2-2 is shown in Fig. 2-3. Here it is clear that the nitrate content varies with time.

2-4 Oxygen Transport

The equation of continuity for gas A in a porous medium can be written as

$$\frac{\partial \epsilon c_A}{\partial t} + \frac{\partial N_{Az}}{\partial z} = R_A \qquad (2\text{-}6)$$

where ϵ = gas porosity, dimensionless

c_A = molar concentration of gas A, moles/L^3

t = time, t

N_{Az} = molar flux of gas A in z direction with respect to stationary coordinates, moles/L^2t

z = distance, L

R_A = molar rate of production of species A, moles/tL^3

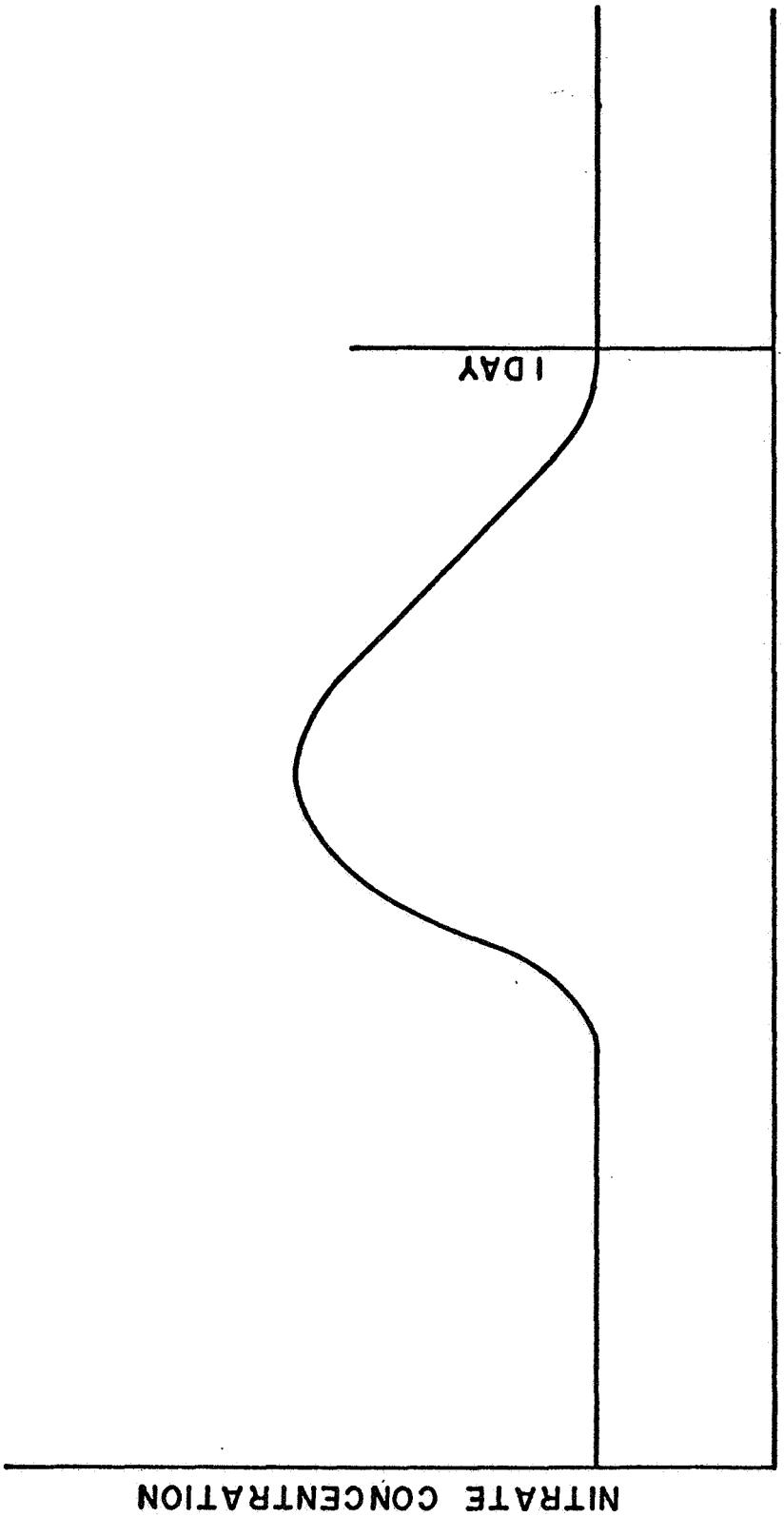


Fig. 2-3 Variation of nitrate concentration in soil water at depth "d".

The molar flux of A can be expressed as the sum of two fluxes, one resulting from bulk flow, the other from diffusion (25). For a three-component system the molar flux of component A is

$$N_{Az} = x_A (N_{Az} + N_{Bz} + N_{Cz}) - c\mathcal{D} \frac{\partial x_A}{\partial z} \qquad (2\text{-}7)$$

in which

x_A = mole fraction of gas A, dimensionless

N_{Bz} = molar flux of gas B in z direction, moles/L^2t

N_{Cz} = molar flux of gas C in z direction, moles/L^2t

c = total molar concentration of gas, moles/L^3

$\mathcal{D}$ = effective diffusivity, L^2/t

The effective diffusivity is less than the diffusivity in free space ($\mathcal{D}_o$) because available cross-section area is reduced and true path length increased by the media. While effective diffusivity depends on factors including pore size and shape, many attempts have been made to relate $\mathcal{D}/\mathcal{D}_o$ to gas porosity only. The resulting empirical or semi-empirical relationships vary with soil type and moisture content. Moisture, aside from serving as a pore filter, can alter pore shape and block some pores. Typically, the relationships are formulated in expressions such as:

$$\mathcal{D}/\mathcal{D}_o = \eta \epsilon^{\sigma} \qquad (2\text{-}8)$$

or

$$\mathcal{D}/\mathcal{D}_o = a\epsilon + b \qquad (2\text{-}9)$$

with η, σ, a, b constants.

Currie (26,27) has fitted the data for several types of porous media with $\sigma = 4$, except at lower porosities, where σ drops below 4. For sand fractions between 0.25 and 0.50 mm, Currie's data can be fitted with $\eta = 16$ and $\sigma = 4$ for ϵ greater than 0.2. For ϵ less than 0.2 the data points fit the expression

$$\mathcal{D}/\mathcal{D}_o = 0.14\epsilon^{1.19} \qquad (2\text{-}10)$$

Equations 2-6 and 2-7 (with suitable effective diffusivity) can be used to study bacterial utilization of oxygen and oxygen transfer in a soil system. For this case, gas A may be oxygen and gas B carbon dioxide. Component C can comprise the remaining gases, including nitrogen, hydrogen and methane. Because oxygen is withdrawn rather than produced, R_A is negative.

One requirement for the solution of equation 2-6 for the transient state is a statement of the initial condition (immediately after the ponded water has completely

infiltrated into the soil). The initial condition is difficult to obtain because of the difficulty of assessing the effect of water infiltration on redistribution of gases in the porous medium. In addition, porosity and rate of oxygen uptake vary with time and distance in some complicated manner during infiltration.

It therefore seems expedient to abandon (at least for now) attempts at describing in mathematical terms the transient-state concentration in the soil and to concentrate on the steady-state form of equation 2-6:

$$\frac{dN_{Az}}{dz} = R_A \qquad (2\text{-}11)$$

True steady state does not occur in a bacterial system unless substrate is continuously added and growth products removed. However, a quasi-steady state, one in which the oxygen concentration depends only on the effective diffusivity and on the bacterial respiration, can exist at some time if temporal changes in these two variables are sufficiently slow.

To solve the quasi steady-state equation for oxygen transport, a model is needed for the variation of bacterial respiration and of effective diffusivity with soil depth.

Another condition that must be specified before the problem can be analyzed is the relationship between

N_{Az} , N_{Bz} , and N_{Cz} . Under aerobic conditions and at steady state, N_{Cz} , the flux of nitrogen, hydrogen, and methane, must be zero because none of these gases are produced in the system.

In their reaeration studies, soil scientists have essentially assumed that $N_{Az}=-N_{Bz}$. This assumption implies that a mole of carbon dioxide is given off for every mole of oxygen used up.

With this assumption, equation 2-7 becomes

$$N_{Az}=-c\mathcal{D}\frac{dx_A}{dz} \qquad (2\text{-}12)$$

and equation 2-11 can be written as

$$\frac{dN_{Az}}{dz}=-\frac{d}{dz}\left(c\mathcal{D}\frac{dx_A}{dz}\right)=R_A \qquad (2\text{-}13)$$

In most cases, however, carbon dioxide production does not equal oxygen consumption. The respiratory quotient, the ratio of moles of carbon dioxide produced to moles of oxygen consumed, can be calculated from the stoichiometry of the equations for the oxidation of carbonaceous matter. The respiratory quotients are 1.0 for oxidation of carbohydrate, about 0.7 for fatty acids, and about 0.8 for proteins. Respiratory quotients greater than unity can occur, for example during synthesis of fats from carbohydrates, but they seldom exceed 1.0 under normal circumstances (28). Respiratory quotients for

nitrification are small negative numbers because about 1/40 mole of carbon dioxide is consumed for every mole of oxygen consumed (See Section 2-2-2).

If $N_{Bz} = -qN_{Az}$ where q is the respiratory coefficient, equation 2-7 becomes

$$N_{Az} = -\frac{1}{(1-x_A+qx_A)}\, c\mathcal{D}\, \frac{dx_A}{dz} \qquad (2\text{-}14)$$

and equation 2-11 becomes

$$\frac{dN_{Az}}{dz} = -\frac{d}{dz}\left[\frac{1}{(1-x_A+qx_A)}\, c\mathcal{D}\, \frac{dx_A}{dz}\right] = R_A \qquad (2\text{-}15)$$

Because the respiratory quotient depends on the substrate and on the oxidation accomplished, its value in intermittent sand filtration is not known, but it is most probably between zero and one. To determine whether the respiratory quotient greatly influences oxygen concentrations, equation 2-11 can be solved with respiratory quotient (q) equal to zero (equation 2-15) and with respiratory quotient equal to one (equation 2-13). A simple model suitable for this purpose is one with constant diffusivity and constant oxygen usage to some depth (L), beyond which activity is zero (Fig. 2-4).

Appropriate boundary conditions are

at z = 0 (surface)	$x_A = x_{AO}$
at z = L	$dx_A/\, dz = 0$

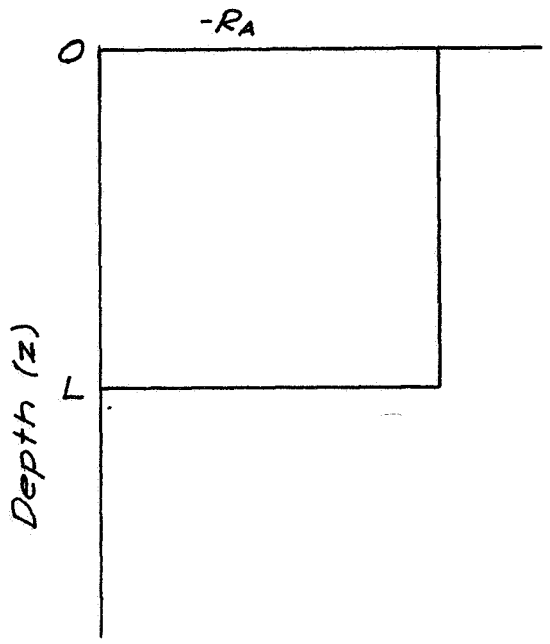


Fig. 2-4 Diagram of profile of bacterial-respiration rates for example to determine effect of respiratory quotient on oxygen profile.

For constant total gas pressure, c, and these boundary conditions, the solution to equation 2-13 is

$$x_A = x_{A0} - \frac{R_A}{c\mathcal{D}} L^2 \left[\frac{1}{2}\left(\frac{z}{L}\right)^2 - \frac{z}{L} \right] \qquad (2\text{-}16)$$

in which x_{A0} is the mole fraction of A at the surface and L is the distance at which activity becomes zero (29,30).

For the same conditions and respiratory quotient of zero, the solution to equation 2-15 is

$$\ln\left(\frac{1-x_{A0}}{1-x_A}\right) = -\frac{R_A}{c\mathcal{D}} L^2 \left[\frac{1}{2}\left(\frac{z}{L}\right)^2 - \frac{z}{L} \right] \qquad (2\text{-}17)$$

A plot of equations 2-16 and 2-17 for two values of $R_A/c\mathcal{D}$ differing by two orders of magnitude shows that the maximum differences in oxygen partial pressures are about 0.015 atmosphere (Fig. 2-5). Because respiratory quotients in intermittent sand filtration are not known and because experimental error may be considerable, a respiratory quotient equal to one may be used in analyzing data.

Equation 2-13 can be used to determine effective diffusivity after x_A and R_A have been determined. For this purpose, equation 2-13 is integrated once with the boundary condition

at $z = L$ $\qquad\qquad$ $dx_A/dz = 0$

and rearranged to yield

$$\mathcal{D}(z) = \frac{\int_z^L R_A(\xi)\,d\xi}{c\,\frac{dx_A}{dz}} \qquad (2\text{-}18)$$

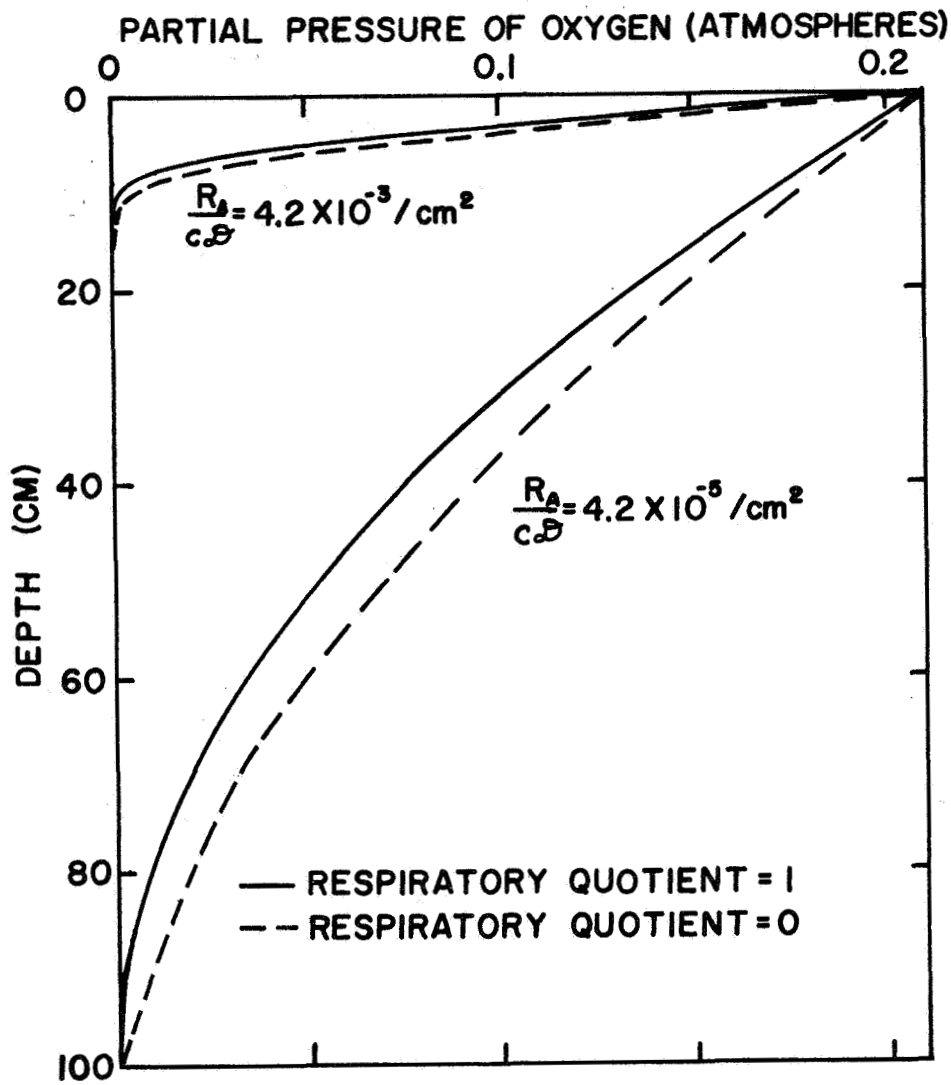


Fig. 2-5 Partial pressures of oxygen for example to determine effect of respiratory quotient on oxygen profile.

in which ξ is a dummy variable and L is the distance at which oxygen concentration is zero. In effect, diffusivity is the flux rate divided by the gradient.

Whenever diffusivity and respiration rates can be estimated with reasonable accuracy, equation 2-13 can be used to calculate the oxygen profile by integrating twice with the additional boundary condition

$$\text{at } z = 0 \text{ (surface)} \qquad x_A = x_{A0}$$

to yield

$$x_A = x_{A0} + \frac{1}{C}\int_0^z \frac{\int_z^L R_A(\xi)\,d\xi}{\mathcal{D}(\xi)}\,d\xi \qquad (2\text{-}19)$$

Because L (the depth at which x_A and, therefore, R_A become zero) is unspecified, trial values of L have to be used in most cases to obtain a solution where both dx_A/dz and x_A are zero at L.

CHAPTER 3

APPARATUS AND ANALYTICAL PROCEDURES

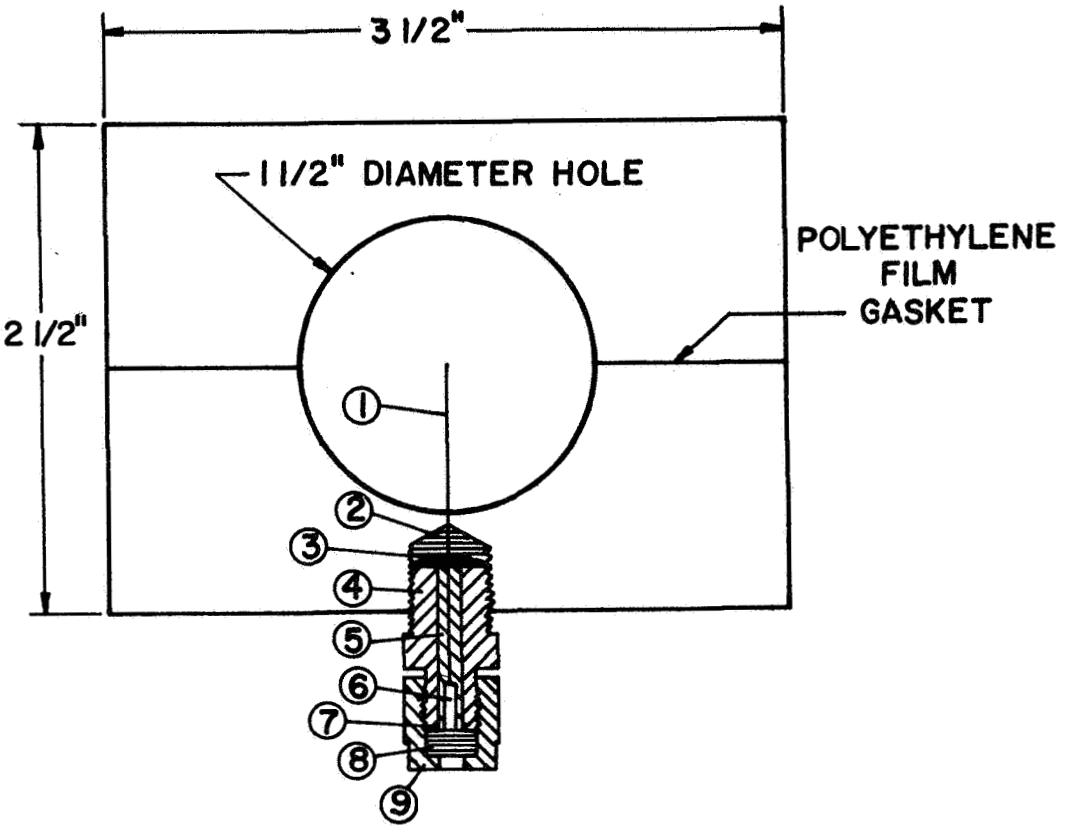
3-1 Soil Columns

Four columns were built so that they could be split axially to remove the soil after they had been operated as laboratory intermittent sand filters. For each column, two acrylic-plastic bars with a cross-section of 1.25 x 3.5 inches were joined to make a combined cross-section of 2.5 x 3.5 inches, and a 1.5-inch diameter hole* was bored axially through the center (Fig. 3-1). Length was sufficient for a 150-cm sand column. The sand was supported by a 200-mesh stainless-steel screen welded to a 0.125-inch length of 1.75-inch O.D. stainless-steel tubing with 0.125-inch wall thickness. A 2-cm layer of fine gravel (4mm) was placed above the sand to distribute the flow and to prevent scouring. A 1.5-inch I.D. acrylic-plastic tube extending above the column allowed for water depth up to 18 inches (Fig. 3-2).

Apparatus for sampling soil atmosphere was similar to that used by Ritchie (31) and Kimball (32). Twenty sampling probes (Fig. 3-1) were placed along each column.

---

* Boring was done by Clark and Wheeler Engineering, Inc., Paramount, California



Legend:

1. 0.032-inch x 0.006-inch wall stainless -steel tube
2. Modeling clay
3. Solder
4. Modified 1/8-inch brass Swagelok male-connector body
5. Brass collar
6. 1/16-inch diameter hole 1/4 inch deep
7. Solder
8. Septum seal for gas chromatograph
9. Swagelok nut

Fig. 3-1 Section of column showing gas-sampling probe.

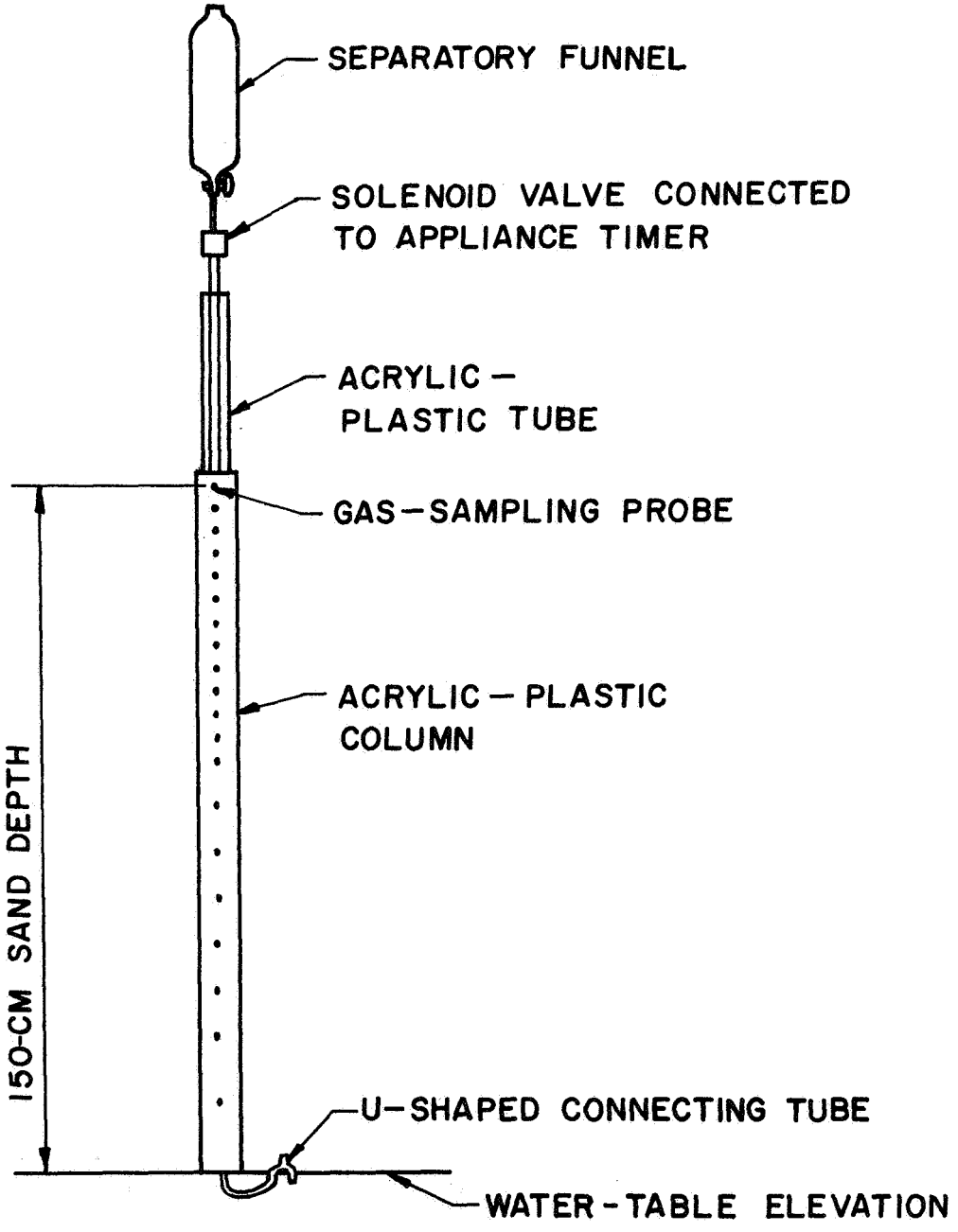


Fig. 3-2 Setup of laboratory columns.

They were located a 5-cm intervals for the first 60 cm of depth and at 10 cm for the second 60 cm. The last one was at 135 cm. Samples were removed by inserting a gas-tight hypodermic needle* through the septum## into the small cavity. Substrate was added periodically to separatory funnels (Fig. 3-2). Flow was automatically regulated with a solenoid valve controlled by an appliance timer. Water-table elevation was controlled with a U-shaped glass connection.

3-2 Apparatus for Gas Sampling in Field

Sampling probes similar to those in the laboratory columns were fabricated to obtain gas samples in field studies. Field probes (Fig. 3-3) were of several lengths for sampling at various depths and contained stainless-steel tubing of larger diameter than the laboratory probes.

Steel rods with a diameter slightly less than the inside diameter of the probes were cut slightly longer than the probe tubing to aid in inserting the sampling probes and to prevent clogging. For sampling, a rod was

---

* Hamilton Company, Inc., Whittier, California. Syringe model 1001LL, needle number KF-72822

Beckman Instruments, Inc., Fullerton, California or The Perkin-Elmer Corporation, Monrovia, California

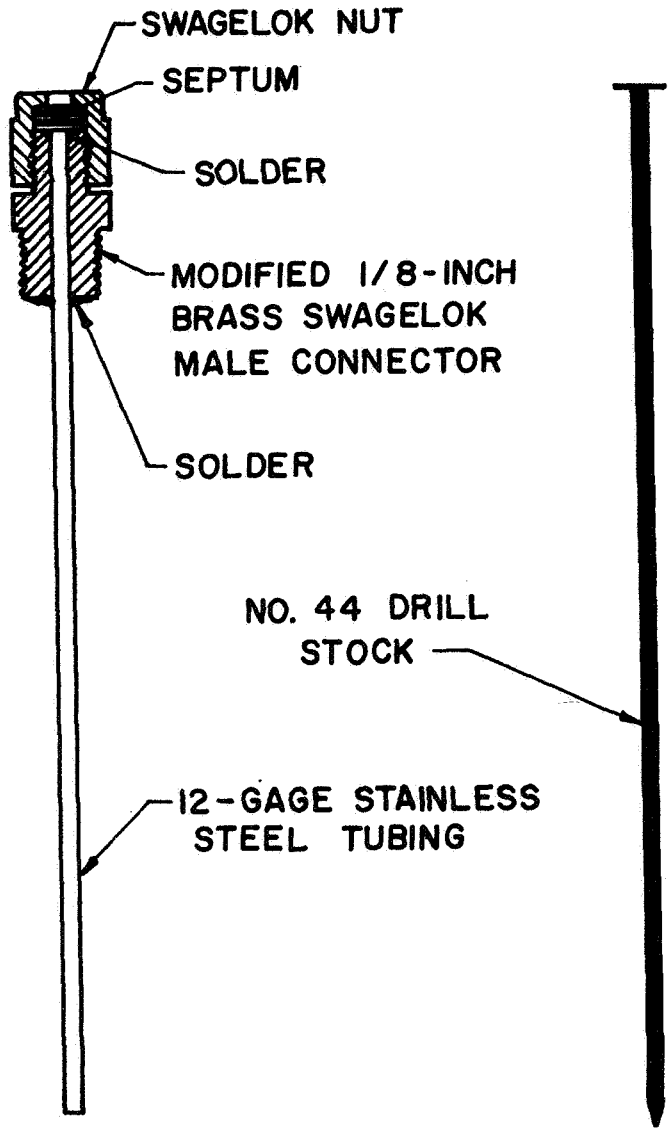


Fig. 3-3 Probe for collecting gas samples in the field and rod to aid in inserting probe into the soil.

placed in the body of a sampling probe and both were inserted vertically into the soil. The rod was removed and the septum and nut were placed on the probe.

Gas was withdrawn by inserting a 30-ml gas-tight hypodermic needle* through the septum. Samples were placed in culture tubes by water displacement (Fig. 3-4). A cap was then screwed on the tube with the tube tip still submerged. A water seal was maintained by placing the capped tube in the inverted position into a water-filled container.

Samples were later withdrawn in the laboratory from the culture tube by reversing the procedure, using a 1-ml gas-tight hypodermic needle.

3-3 Gamma-Radiation Equipment

Soil moisture determinations were obtained by using a 100-millicurie cesium-137 source and measuring gamma-radiation attenuation. For these measurements a soil column was placed in a rack that allowed the column to move vertically between source and detector (Fig. 3-5). The shielding was patterned after that of Davidson, Biggar, and Nielsen (33).

The Cs^{137} source was contained in a hole machined lengthwise through the center of a 4 x 4 x 8-inch lead

---

* Hamilton Company, Inc., Whittier, California. Syringe model 1030LL, needle number KF-72822.

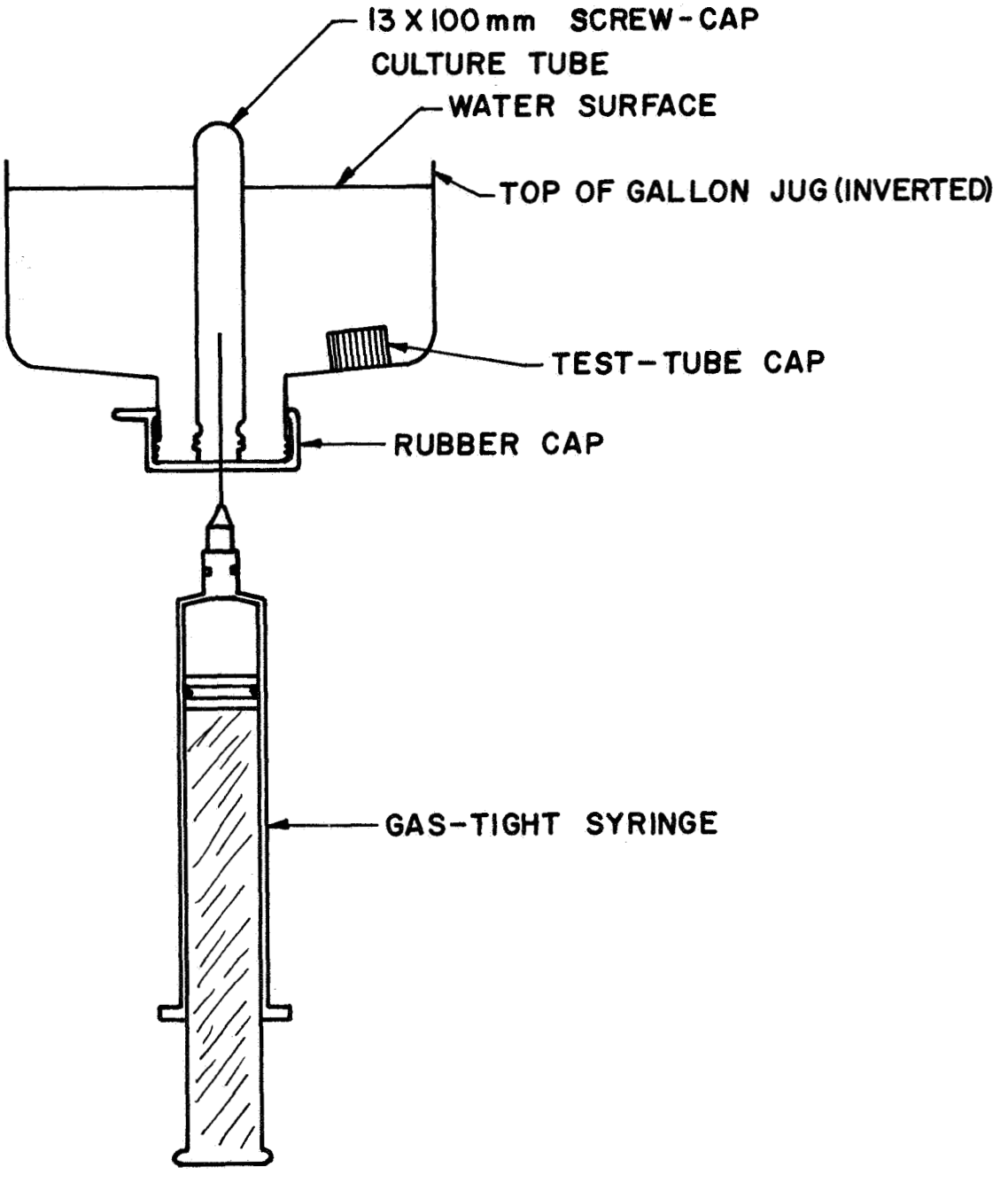


Fig. 3-4 Apparatus to contain gas samples collected in the field.

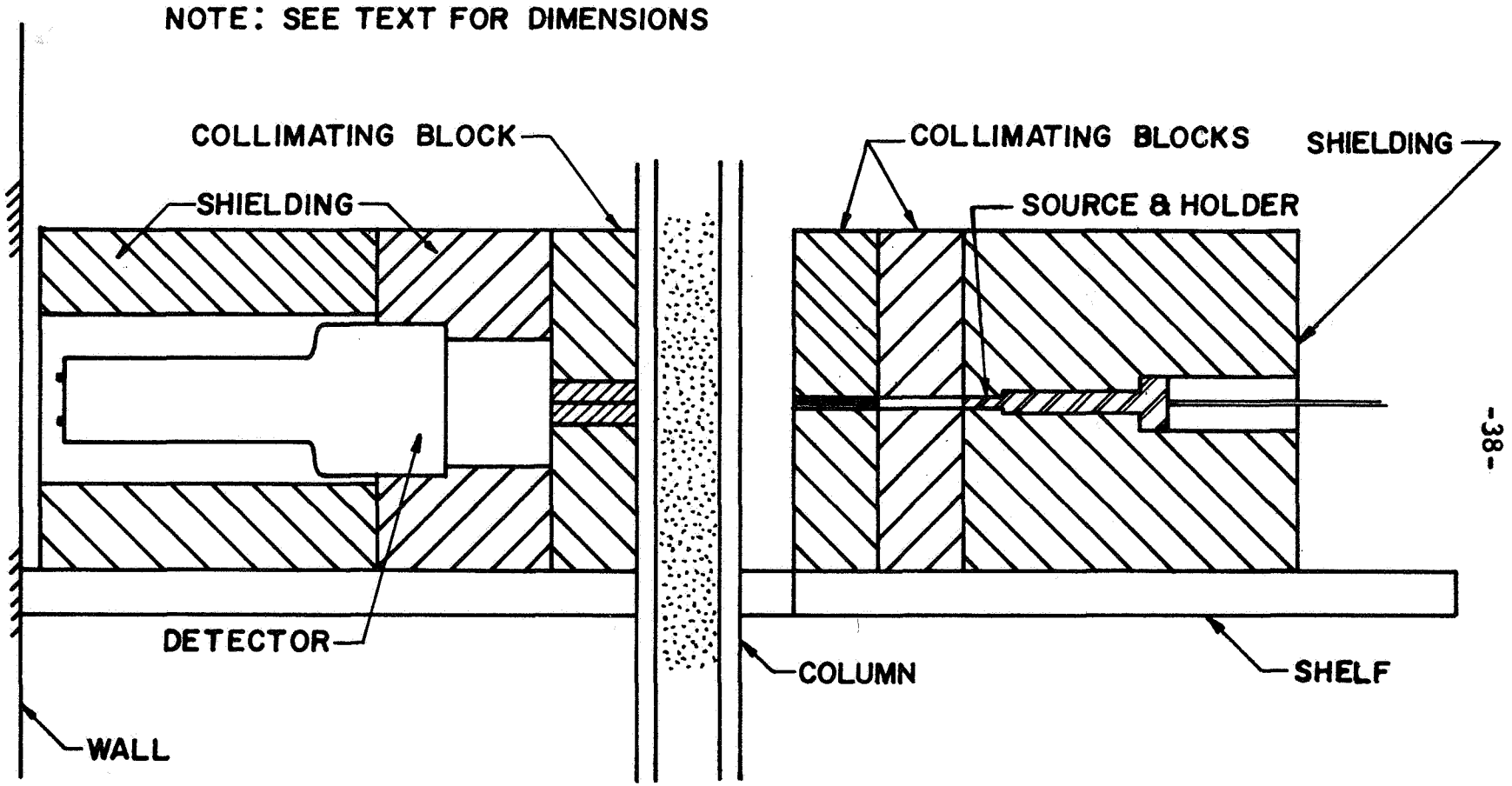


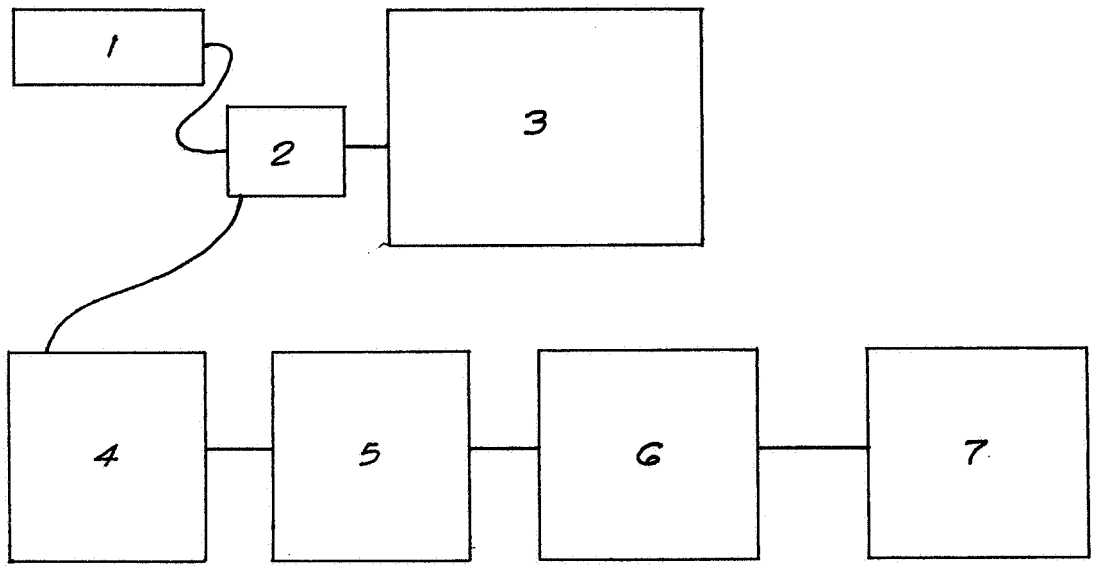
Fig. 3-5 Detector, column, and source used to measure moisture by gamma-ray attenuation.

brick. Six 2 x 4 x 8-inch bricks surrounded the brick containing the source, forming an 8 x 8 x 8-inch container. Radiation from the source was collimated before reaching the soil column by passing through openings in two lead bricks. The brick nearest the source was 8 x 8 x 2 inches, with a 0.250-inch diameter hole, 2 inches long, through the center. The second brick was 8 x 4 x 2 inches, with 8-inch side vertical, and contained a 2-inch stainless-steel tube having inside diameter of 0.125 inches and an outside diameter of 0.250 inches.

The front section of a scintillation detector was inserted in the center of a 4 x 8 x 8-inch lead block, with the front of the thallium-activated sodium iodide crystal flush with the front of the block. The face of the container for the detector was covered with an 8 x 8 x 2-inch plate containing a .875-inch diameter lead plug drilled to accept a 2-inch-long stainless-steel tube with inside diameter of 0.062 inches and outside diameter of 0.083 inches.

Several collimating arrangements were tried before the one described was selected.

Counting equipment to measure gamma-radiation attenuation by the empty columns and to check consistency of packing is shown in Fig. 3-6. The ratemeter signal is for a 10 mv (millivolt) recorder.



Legend:

1. Scintillation detector (Harshaw Type 125/QGX)
2. Preamplifier
3. Power supply (John Fluke Manufacturing Co, Inc., Model 409 A)
4. Single-channel analyzer (Baird Atomic Model 250)
5. Scaler (Nuclear-Chicago Model 192 A)
6. Ratemeter (Nuclear-Chicago Model 1620 B)
7. Recorder (Nuclear-Chicago R-2000 series or Sargent Model SR)

Fig 3-6 Schematic diagram of γ-radiation-counting apparatus

Because differences in gamma-radiation attenuation during drainage were less than ten percent of attenuation after drainage, it was desirable to expand the portion of the recorder scale showing the differences. A recorder (Sargent Model SR) allowing a variation in range was used in tests involving soil-moisture movements. The scale zero was suppressed by an apparatus that supplied a small voltage to resist the ratemeter signal. The apparatus consisted of a 50,000-ohm potentiometer in parallel with a Heath Company pH-MV Test Unit. The resulting voltage was changed by varying the potentiometer settings or voltage settings on the test unit. The 1-mv recorder scale was used when the ratemeter signal was suppressed.

3-4 Porous Media

Ottawa quartz sand was one of the porous media used in the columns. When clean, it had a geometric mean diameter of 0.56 mm, a geometric standard deviation of 1.2, a density of 2.61 gms/cc, and a porosity of 0.35. An abundant microbial culture was grown on the sand by placing the sand in temporary columns and dosing intermittently with settled sewage.

Soil from the top six inches of the Whittier Narrows Test Basin was used in one column. The soil and operating history of this test basin are described by McMichael and McKee (13).

3-5 Chemical Analyses

Ammonia nitrogen was determined by direct Nesslerization (34). A sample containing less than 0.25 mg of ammonia nitrogen was placed in a 100-ml Nessler tube. To this sample 0.5 ml of 10 percent sodium hexametaphosphate was added to prevent deposition of magnesium and calcium salts upon addition of 2 ml of Nessler reagent (35). Absorbance at 420 mμ was read and compared with standards.

Kjeldahl-nitrogen determination for organic nitrogen plus ammonia was performed by a micro-analytical method using a mercuric sulfate catalyst with final ammonia determination by Nesslerization. The method is similar to that in Standard Methods (35). The reagent volumes have been adjusted for 50-ml Kjeldahl flasks instead of 800-ml flasks.

Nitrate and nitrite determinations were performed according to Standard Methods (35). The brucine method was used for nitrate and a modified Griess-Ilosvay procedure for nitrite. Extraction from soil water was by a method similar to that of Bremner (36). Twenty-five grams of soil were placed with a saturated calcium sulfate solution into a 250-ml Erlenmeyer flask. Seventy-five ml of calcium sulfate solution were used for tests with Ottawa sand; samples from the Whittier Narrows test basin

were added to 125 ml. The flask was stoppered and shaken for 10 minutes on a mechanical shaker and the suspension settled. The supernatant was decanted and filtered through a Whatman No. 42 filter paper until the filtrate was clear. The filtrate was then analyzed. To prevent further nitrification the samples were kept in a cold room at 40° F between steps.

The analytical method for chemical oxygen demand was the Standard Methods technique (35).

Glucose and carbohydrate determination was colorimetrically by the anthrone method (37). Except for furfural, no non-carbohydrates have been found to yield positive results with anthrone reagent (37). Of the carbohydrates, the hexoses seem to produce the most intense color. Glucose and fructose produce about equal intensity and galactose yields 54 percent the color of glucose. The contribution from pentoses and uronic acids are negligible at the wavelength (625 $m\mu$) and the anthrone concentration used (38). Carbohydrate was determined in soil samples by placing 5-gram samples and 50 ml of 3N H_2SO_4 in 125-ml Erlenmeyer flasks with mouths covered by marbles to reduce evaporation. The samples were hydrolyzed by placing the flasks on a steam bath for 24 hours. Hot hydrolyzate was passed through medium fritted-disk filters and the residue washed with 50 ml of

water (38). Samples were diluted and cooled before chemical determination by the anthrone method. Carbohydrate was expressed as "glucose equivalent."

3-6 Gas Analysis for Oxygen

Gas analyses for oxygen were made polarographically with an oxygen electrode built by Radiometer of Copenhagen, Denmark. The microelectrode comprises a platinum cathode and silver/silver chloride cathode placed in an electrolytic solution behind a teflon membrane permeable only to gases. The unit is enclosed in a thermostated cell at 38° C. A sodium sulfite solution was used to zero the instrument; air was used to set its range.

Small gas volumes were removed from the columns using the probes described above. A gas-tight syringe needle inserted through the sampling-probe septum withdrew a 1-ml volume which was inserted into the constant temperature cell of the oxygen electrode.

3-7 Measurement of Respiration Rates

Respiration rates were determined manometrically with an American Instrument Company Rotary Warburg apparatus. The constant temperature bath was controlled at 25.35° ± 0.02° C by a mercury thermoregulator. Manometer fluid was prepared from Spec. D-2930 Meriam Indicating Fluid Concentrate. (0.998 specific gravity at 25° C)

The center wells of 125-ml BOD Warburg flasks were greased, after which approximately 25 grams of sand were added. One ml of 10-percent KOH and a folded filter paper were placed in each well. Flasks were attached to manometers and placed in the bath, with stopcocks open. After five minutes of shaking, joints were adjusted. The flasks were allowed to equilibrate for 10 to 15 minutes. Manometer fluid was adjusted to reference points, stopcocks were closed, and readings were begun.

Exact sand weights were determined after the measurements were complete and flask constants calculated (39).

Three thermobarometers containing sterile sand were used to correct for temperature or pressure variations.

3-8 Soil-Moisture Determination

3-8-1 Gravimetric Determination

A weighed sample was dried for 48 hours at 100 to 110° C and cooled in a desiccator before reweighing.

3-8-2 Moisture-Retention Measurements

A method described by Vomocil (40) was used to determine moisture retention above a water table. The apparatus consisted of a 60-ml Buchner funnel (ultra-fine fritted glass) connected to the bottom of a 50-ml burette by 1/16-inch tygon tubing. After the funnel was boiled to remove air from the disk, the funnel below the disk, the tube, and the burette were filled with water.

Sand was placed in the funnel and water added to submerge the sample for 24 hours. Excess water was drained off and loose covers placed on the funnel and burette to reduce evaporation. The burette was lowered in small intervals and the soil water allowed to flow through the disk into the burette. After drainage had ceased, the burette reading and elevation difference between the sand and the water surface in the burette were recorded. The burette was then lowered again and the process repeated several times.

After these measurements, the moisture remaining in the sand was determined as in section 3-8-1.

3-8-3 Measuring Soil Moisture by Gamma-Ray Attenuation

Moisture-content measurements are based on γ -radiation attenuation by soil water.

The attenuation equation for monoenergetic radiation is

$$I = I_o \exp(-\mu \rho z) \qquad (3\text{-}1)$$

where I = measured radiation with interference

I_o = measured radiation with no interference

μ = mass absorption coefficient of absorbing material for the given energy of radiation (L^2/M)

ρ = density of material (M/L^3)

z = sample thickness (L)

Since μ varies with radiation energy, equation 3-1 is strictly correct only for radiation of a given energy. This condition was approximated by using an analyzer that measured only radiation with an energy within a narrow range.

The energy spectrum for Cs^{137} has a radiation peak at 0.66 Mev. (Fig. 3-7). The abscissa does not represent actual energy values of the γ photons. The values are, however, proportional. While it would be desirable to measure only those photons with an energy very near the peak, such a limitation is impractical because the position changes slightly with density changes and because instrument amplification drifts slightly. Effects caused by changes in peak position can be minimized by setting upper and lower energy limits symmetrically about the peak so that the slope at the lower setting is zero. For Fig. 3-7 these settings would be 44 and 62 volts.

Equation 3-1 can be rewritten as

$$I = I_o \exp -(\mu_w \rho'_w z_w + \mu_s \rho'_s z_s + \mu_p \rho_p z_p) \qquad (3\text{-}2)$$

in which subscripts refer to water, sand, and plastic. Primes are used for water and sand densities to indicate that these are bulk densities because the substances do not occupy the entire space denoted by z. The equation indicates that moisture content can be determined by measuring changes in attenuation caused by moisture since

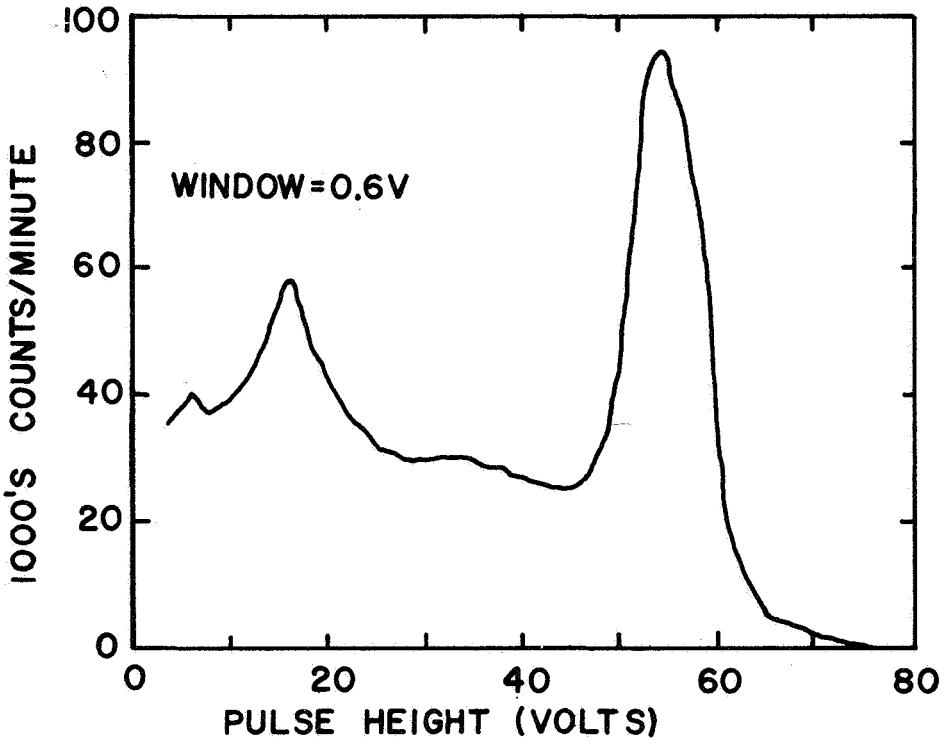


Fig. 3-7 Pulse height spectrum for Cs^{137}.

terms relating to container and sand are constant.

All the terms enclosed by parentheses in equation 3-2 remained constant during the experiment, except for ρ'_w . Hence, the equation can be rewritten as

$$I = A\exp(-\mu_w \rho'_w z_w) = A \exp(-\mu_w \rho_w \theta z_w) \qquad (3\text{-}3)$$

where $A = I_o \exp-(\mu_s \rho'_s z_s + \mu_p \rho_p z_p)$

The value $\mu_w \rho_w z_w$ was obtained by measuring the attenuation of water in a special rectangular container having a 1-1/2-inch hole bored through it. The term θ was calculated from measurements on columns in operation.

CHAPTER 4

EXPERIMENTAL PROCEDURES

4-1 Field Experiments

Field studies were conducted at the Whittier Narrows Test Basin used in studies by McMichael and McKee (13). The basin, built on loamy soil previously used for agriculture, is 50 ft by 70 ft in plan and is enclosed by two-ft high levees. It is equipped with sampling facilities consisting of a central well with four sampling pans at depths of 2, 4, 6, and 8 ft below the ground surface. The two-ft diameter pans are spaced 60 degrees apart 15 ft from the center of the well and drain to the well through 3/4-inch ID vinyl tubing. Depth to the water table is about nine ft. The basin is covered with a six-inch layer of pea gravel to reduce weed growth.

The Los Angeles County Flood Control District spreads 1.4 to 2.0 ft of activated-sludge effluent from the Whittier Narrows Wastewater Reclamation Plant on the basin once a day, five days a week. Infiltration takes from 10 to 14 hour.

4-1-1 Analysis of Soil Water

Soil borings to depths of approximately eight feet were taken on different days beginning 1/2, 5, and 14 hours after the waste had completed its infiltration. A soil boring about one foot long was also taken about six hours after the basin had been spread when the remaining water

depth was approximately one foot. Boring operations generally took about one hour. Soil samples were brought to the laboratory (about 11 miles distant) where nitrate in the soil water was determined.

Infiltration took about 10 hours when these studies were made.

4-1-2 Analysis of Percolate

Incremental samples of percolate were collected at the Whittier Narrows Test basin for 14 hours after the basin was dosed to a depth of 1.76 ft with effluent. Samples of about 100 ml and 150 ml each were collected from the two-ft pan and 250- and 500-ml samples from the six-ft pan.

The samples were brought to the Whittier Narrows Water Reclamation Plant at intervals of from one to two hours and analyzed immediately for nitrates. Unused portions were then preserved by acidifying and analyzed for Kjeldahl nitrogen later in the week. Sufficient numbers of these samples were analyzed to obtain trends with time for nitrate and Kjeldahl nitrogen.

4-1-3 Gas Collection and Analysis

Gas samples were collected at depths ranging from 5.25 inches to 8 ft for times after completion of infiltration of from 5 to 46 hours. Infiltration took about 14 hours when these samples were taken.

A rod was placed in the body of a field probe (Fig. 3-3) and both were pushed into the soil until the end of the probe was at the desired depth. The rod was then removed and the septum and nut replaced on the probe.

Samples were collected by a 30-ml gas-tight hypodermic needle inserted through the septum. The syringe plunger was pulled out to the desired volume and held for one minute before the needle was withdrawn. The first sample from a probe location, about three times the probe volume, purged the tube and was discarded. About 10 ml gas volumes were then withdrawn from the probe and collected in test tubes.

4-2 Laboratory Experiments

4-2-1 Filling Columns

The gamma-ray attenuation by the empty columns was measured before filling by taking counts of approximately 100,000 at four locations in each column. Attenuations for individual columns were in close agreement. The largest error found was 1.4 percent from a column mean.

Sand was removed from the temporary columns, mixed thoroughly, air-dried overnight and carefully packed into columns. Soil from the Whittier Narrows Test Basin dried in clumps which were pulverized before being packed. One person poured sand in increments of approximately 50 ml while a second tapped the column at the sand level with a plastic mallet.

Seven fillings were made during the tests. The uniformity for the first six was checked by measurements of gamma-ray attenuation. Measurements for individual columns were always within 1.6 percent of the mean.

4-2-2 Operation of Columns

Four columns (labelled 1 through 4) were initially filled with 0.56-mm Ottawa sand well seeded with settled sewage and subjected to identical loading patterns but different substrates. Because the solids in wastewaters applied to intermittent sand filters are mostly dissolved, only soluble substrates were first used. Substrate consisted of glucose, ammonium chloride, and salts. Columns 1 and 2 received 400 and 150 mg/1, respectively, of ammonium chloride and no glucose. Column 3 received 200 mg/1 glucose and 42 gm/1 ammonium chloride and Column 4 received 100 mg/1 glucose and 21 mg/1 ammonium chloride. Gas analyses taken six weeks after operation had begun indicated little change in oxygen tension in the columns. At that time, substrates to Columns 1 and 3 were strengthened, the medium in Column 2 was changed to soil from the Whittier Narrows Test Basin, and the sand in Column 4 was changed to one composed of about one-half that previously in the column and one-half sand with geometric mean diameter of 0.16 mm and geometric standard deviation of 1.8.

Substrates and loading patterns for Columns 1 and 3 were changed periodically in an attempt to minimize time required to obtain a measurable oxygen gradient in the column atmosphere. Table 4-1 summarizes the operation of these two columns by listing the substrate concentrations and volumes added at the beginning and at the end of the tests and also the limits of their variations. Infiltration times for dosages of 500 ml were from 2 to 12 hours (Column 1) and 1 to 3 hours (Column 3), depending on the condition of the sand surface. Scarifying the surface down to five centimeters reduced infiltration times toward the lower figures. Column 3 was operated for 4 months. Studies on Column 1 were conducted over about 5-1/2 months.

The Whittier Narrows soil in Column 2 drained slowly after addition of secondary effluent and formed clumps about 1/4 inch in diameter. Only a few air measurements were taken.

Column 4 did not allow sufficient flow with the sand mixture and was repacked with 0.56 mm Ottawa sand. The column was dosed almost daily for 2 months with 500 ml of settled sewage passed through glass wool to reduce suspended matter. Settled sewage was obtained from the Whittier Narrows Water Reclamation Plant and refrigerated until used.

TABLE 4-1

Summary of Column Operation

	Column 1			Column 3		
	Initial	Final	Range	Initial	Final	Range
Volume/dose (ml)	250	500	--	250	500	--
Depth/dose (cm)	22	44	--	22	44	--
Doses/day	1	1	1-2	1	2	--
Constituents						
Glucose (mg/1)	0	250	0-500	200	500	--
NH_4Cl (mg/1)	400	100	--	42	100	42-210
1.0 M Potassium phosphate buffer pH 7 (ml/1)	1	10	--	1	10	--
$MgSO_4 \cdot 7\ H_2O$	20 mg/1					
$FeCl_3 \cdot 6\ H_2O$	0.5 mg/1					
$MnSO_4 \cdot H_2O$	10 mg/1					
$CaCl_2$	7.5 mg/1					
Tap Water	50 mg/1					

same for all tests

4-2-3 Gas Analyses

Gas samples were taken at various times after dosing to determine the variation in oxygen profile during a loading cycle.

The preliminary plan was to obtain only one profile measurement per cycle to avoid distorting air movements. Because measurements were not reproducible from cycle to cycle, owing to changes in surface condition, it was necessary to obtain sequences of measurements during single cycles.

Some probes plugged after a time and could no longer be used for sampling. The probes in Column 1 were unplugged by removing the Swagelok nuts and septa and passing a piano wire through the fine tube.

4-2-4 Effluent Analyses

Fifteen-ml increments of effluent were collected to obtain data relating composition and throughput. Analyses included Kjeldahl nitrogen, glucose, nitrate and nitrite nitrogen, and chemical oxygen demand.

4-2-5 Measuring Respiration Rates

Following column experiments, Column 3 was disassembled to measure oxygen uptake rates. Five-hundred ml of substrate were added to the surface and the column allowed to drain until more than 475 ml of effluent had been collected. Infiltration took about one hour and

drainage about three hours.

The column was then disassembled and soil samples from various depths were placed in Warburg flasks (see Section 3-7) for determination of respiration rates. Oxygen uptake measurements were begun five hours after the column was dosed and continued until 76 hours after addition of nutrient.

4-2-6 Moisture Measurements

Moisture determinations by gamma-attenuation measurements were made at a few depths on Column 3. The column was placed on the rack for radiation measurements about one-half hour before substrate was added and radiation passing through the column was traced with the recorder range at 10 mv. The scale zero was then suppressed until recorder readings were about 10 percent of full scale. The scale was next expanded by changing the recorder range to 1 mv for moisture movements. After addition of substrate to the column, recording was continued until attenuation became almost constant.

Fifteen sand samples were taken after disassembly of Column 3 to obtain moisture content gravimetrically. Moisture-retention measurements were also made on clean sand to determine the effect of bacterial growth on moisture retention.

4-2-7 Chemical Analyses of Sand

Sand from various depths was taken from Columns 3 and 4 after disassembly. Sand from Column 3 was analyzed for chemical oxygen demand, and carbohydrate (glucose equivalent). Samples from Column 4 were analyzed for nitrate and nitrite.

CHAPTER 5

RESULTS AND DISCUSSION

5-1 Field Experiments

5-1-1 Analysis of Soil Water

Results of analyses for nitrates in soil water at Whittier Narrows are shown in Fig. 5-1. The diagrams indicate nitrate concentrations in soil water about 1/2, 5, and 14 hours after water had infiltrated, and also when the surface was still ponded.

Although the data show considerable scatter, the results during ponding and shortly after infiltration is completed tend to support the hypothesis presented previously that bacterial activity is limited while the surface is ponded and that nitrate content is initially constant with depth after the surface of a soil basin has just drained. The figure also suggests that nitrifying activity progresses downward into the soil as oxygen enters into the soil.

Samples taken 1/2 hour after infiltration show that nitrification has already taken place near the surface and indeed down to one foot or so. After 5 and 14 hours, nitrification has progressed down to about two and four feet, respectively. After 14 hours, the nitrate concentration is about 40 mg/1 for the top four feet and about 15 mg/1 below this depth. Note that the nitrate content of the ponded water was about 6 mg/1.

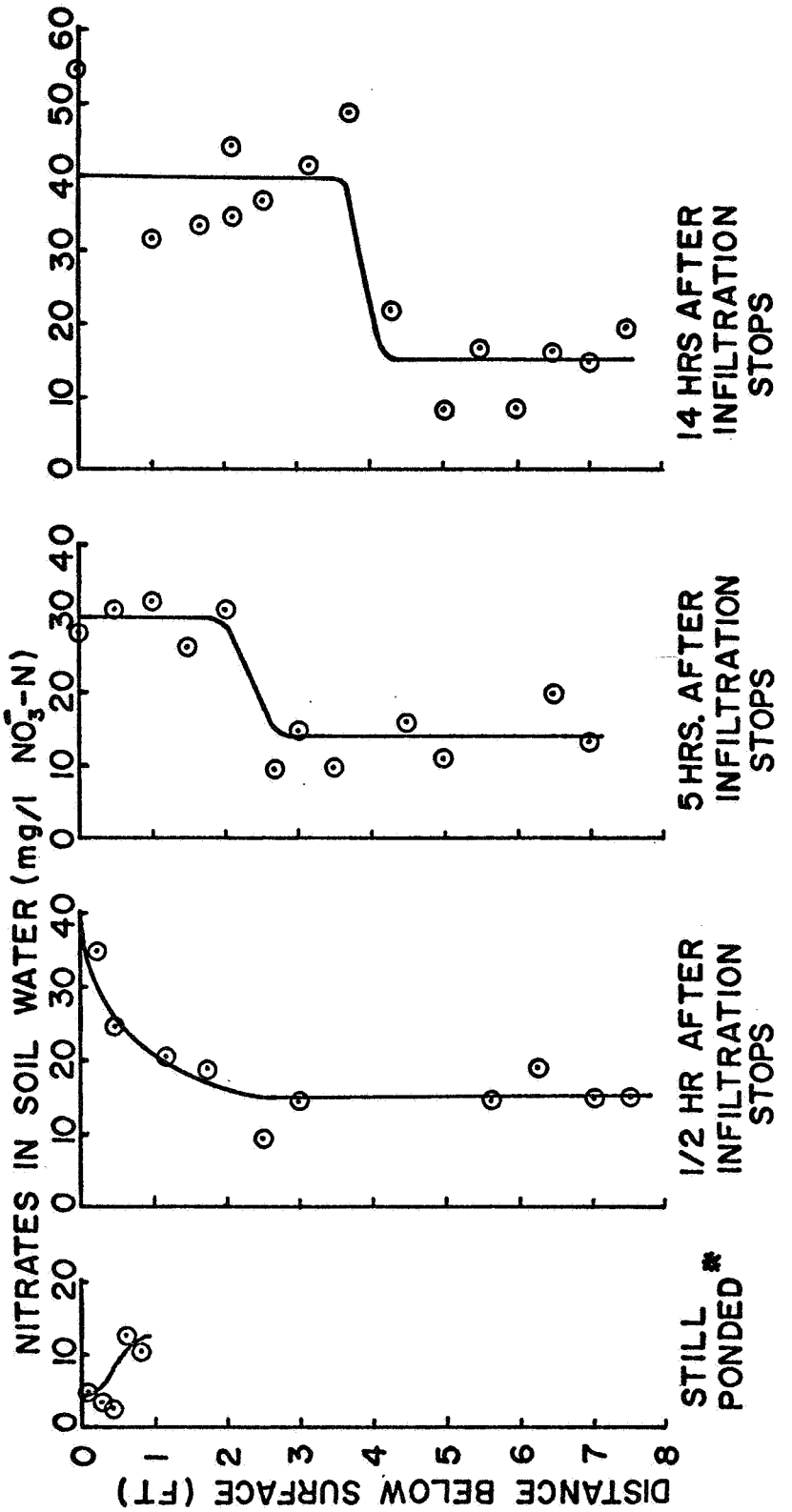


Fig. 5-1 Nitrate profiles in soil water at Whittier Narrows Test Basin. (Ten-hour infiltration time)

No attempt was made to measure ammonium or other forms of nitrogen with minus-three oxidation state. These tests were to measure only dissolved substances containing nitrogen. Extraction as used in determining nitrate in the soil water would have removed ammonium weakly adsorbed to soil surfaces and caused nitrogenous matter to slough off during shaking.

The model presented in Section 2-3 proposed that nitrate below four feet is low because the liquid passed through the biologically active zone when oxygen was low and therefore was never nitrified. An alternative hypothesis for the decrease in nitrate below four feet could be that denitrification has taken place. However, data from the report by McMichael and McKee (13) for samples at six and eight feet indicate no decrease in total nitrogen. Discussion in section 5-1-3 presents further evidence against denitrification.

5-1-2 Analysis of Percolate

Incremental samples collected from the sampling pans did not demonstrate the nitrate wave sketched in Fig. 2-3. Kjeldahl-nitrogen concentrations for samples from the pan at a depth of two ft remained at about 0.5 mg/l through the entire sampling period;from the six-ft pan at about 1.8 to 2.0 mg/l. Concentrations of nitrate nitrogen for effluent taken from the two-ft pan varied from about 24

to 32 mg/l after the initial few hundred ml in which nitrate was somewhat lower. Effluent from the six-ft pan was not analyzed for nitrate.

Using a sodium chloride tracer on the same basin, McMichael (41) showed that there was a 38-hour delay between the time tracer was added to the ponded water and the time it was first observed at the two-ft pan. The delay was 28 hours at the six-ft pan. In addition, the quantities of water collected were only 8 and 17 percent of those expected and the flow-through characteristics of the four sampling pans were markedly different. These results imply that the water collected at the pans is not representative of newly percolated water.

Water perched in the capillary fringe above pans impedes flow into the pans and diverts flow around them. Thus, the composition of water collected by sampling pans does not represent that of water percolating nearby, but rather that of water held in the fringe for a day or more.

Problems caused by perched water can be avoided by using porous ceramic cups through which soil solution is extracted by applying tension to the cups with a vacuum pump or hanging water column (42, 43). However, a better way to obtain a continuous sample of percolate that is truly representative seems to be by means of column experiments, in which case the entire percolate is collected.

In addition, the column experiments can be used to evaluate the proposed model.

5-1-3 Gas Collection and Analysis

The data for oxygen content of gas samples taken at various depths at Whittier Narrows for times of 5 to 46 hours after completion of infiltration are plotted in Fig. 5-2. Each point is the average of two samples taken at a probe location. Samples with differences of greater than 10 mm mercury (.013 atmospheres) were not plotted. Samples could not be obtained from some locations, possibly because the probes became plugged with mud as a partial vacuum was applied by withdrawing gas.

The data show no trend with time but are scattered within the envelopes in Fig. 5-2. A small part of the scatter may be due to leakage around the septa, but most irregularity was probably caused by the non-uniformity of the soil porosity. In sampling of this type, the sample is drawn from the larger pores about the probe. Hence any cracks about the bottom of the probe would have contributed a disproportionate part of the gas volume and the sample withdrawn would not have given a true indication of the oxygen content at the sampling point.

The results indicate that oxygen did not penetrate even to two feet at the test basin during these tests.

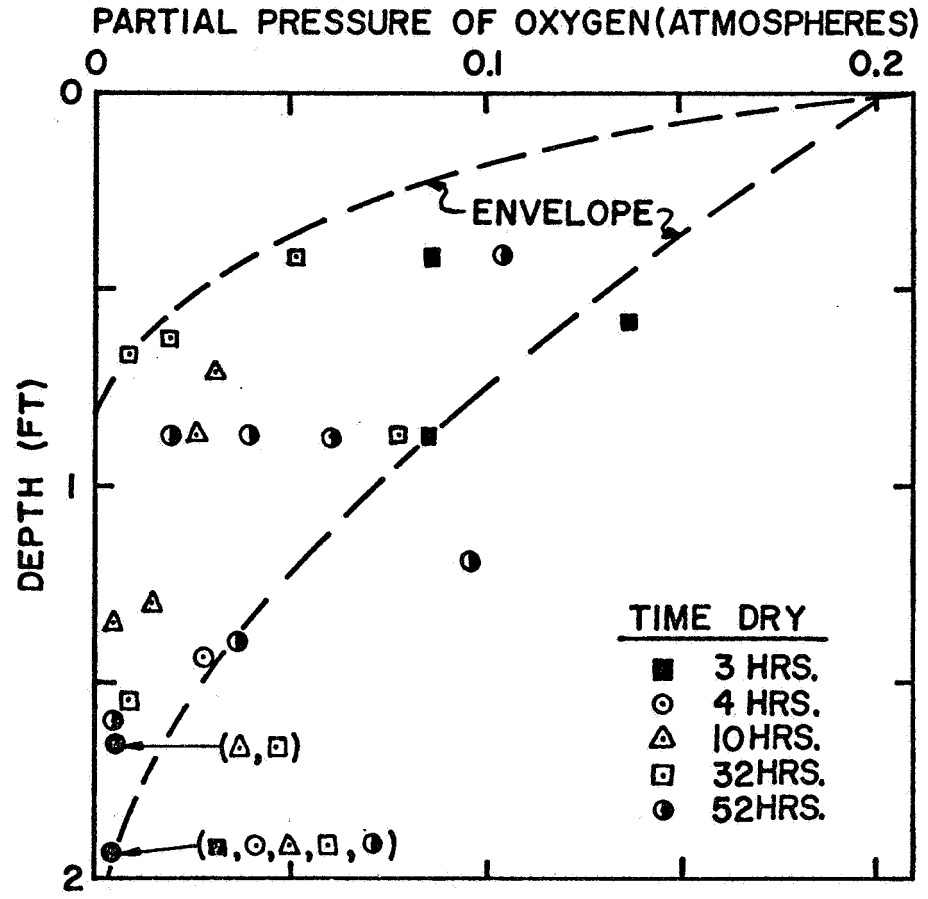
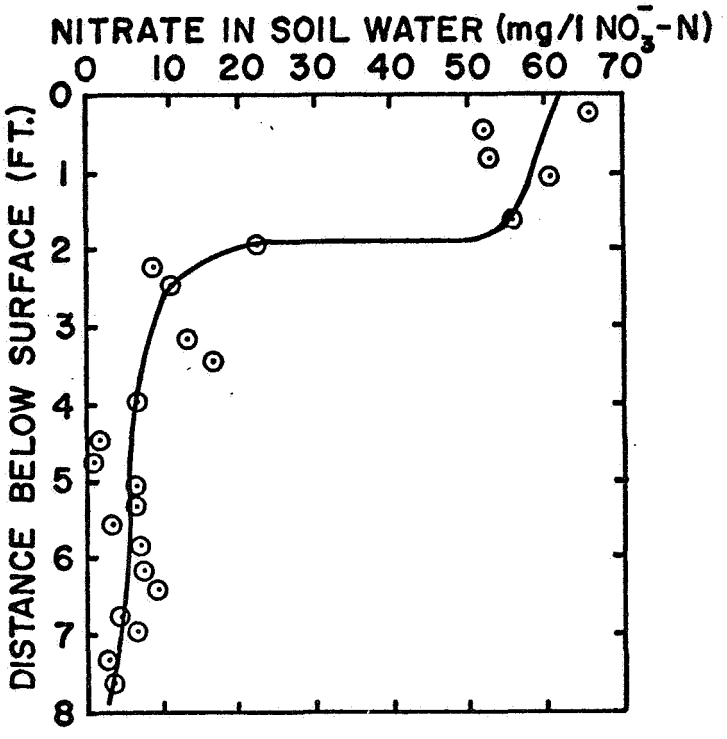


Fig. 5-2 Measurements of partial pressure of oxygen in soil atmosphere at Whittier Narrows Test Basin.

The gas samples were taken about nine months after the cores for analysis of soil water were taken, during which interval the infiltration time increased from 10 to 14 hours. A new core was thus taken to determine whether the nitrate profile had changed. The results for a core taken about 11 hours after completion of infiltration (Fig. 5-3) show that the active zone for nitrification has decreased to less than two feet, roughly the depth where anaerobic conditions begin (Fig. 5-2).

In section 5-1-1, denitrification was presented as a remote possibility for the decrease in nitrate content beyond the first few feet of an intermittent sand filter. Denitrification could occur, of course, only if there had been prior nitrification. Fig. 5-1 shows that nitrate content was low for all depths immediately following infiltration. Data in Fig. 5-2 show that oxygen did not penetrate more than two feet into the soil when the data for Fig. 5-3 were collected, so that nitrification could not have taken place beyond two feet at that time (or beyond four feet when the data for Fig. 5-1 were obtained). Since nitrification was absent, denitrification could not have caused the large decrease in nitrate concentration.



5-3 Nitrate profile in soil water at Whitter Narrows Test Basin 11hours after completion of infiltration. (Fourteen-hour infiltration time.)

5-2 Laboratory Experiments

5-2-1 Gas Analyses

Oxygen profiles in the laboratory columns were not reproducible from cycle to cycle, but depended on the condition of the sand near the surface, where bacterial growth was observed to be much more dense than elsewhere. The growth reduced infiltration rates and effective gas diffusivity. Because short infiltration times were wanted to allow atmospheric oxygen to be available for a large part of the cycle, the column surface was scarified from time to time to increase infiltration rate. Besides increasing infiltration rate, scarifying also allowed oxygen to penetrate to a greater depth.

The results of oxygen analyses for gas samples taken during two cycles of Column 3 are shown in Fig. 5-4. The samples for each cycle were taken at 7.9, 24.5, 49.3, and 71.3 hours after substrate was added. As noted, the profiles are not reproducible, but the two sets of data are similar in shape and trend. Because the curves for each set do not vary greatly over a time span of more than 63 hours, the assumption that the system reaches a quasi-steady state after some time seems tenable. Such reasoning would be poor if the curves in a cycle differed greatly. With rapid changes, oxygen concentrations would depend not only on conditions (such as effective diffusivity

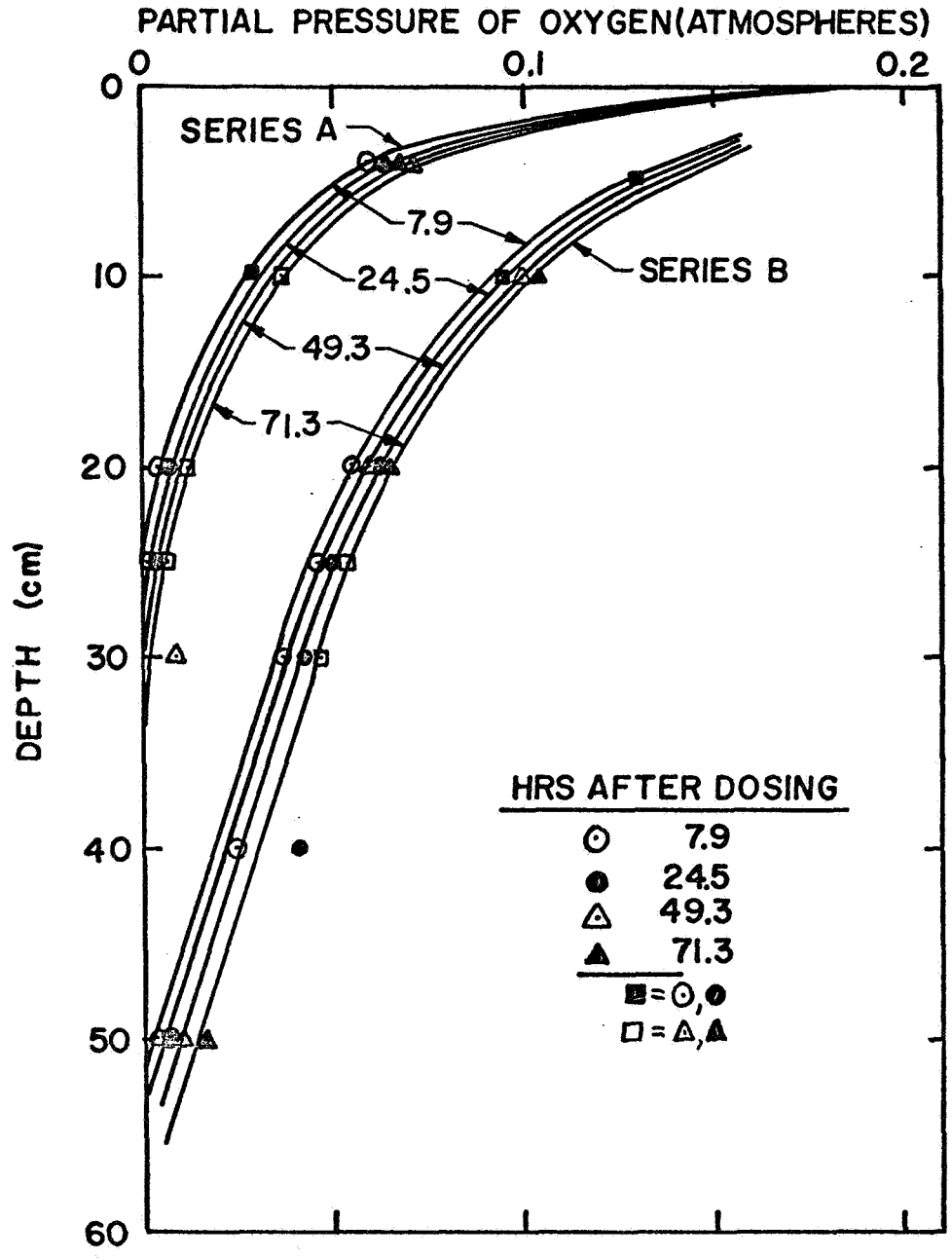


Fig. 5-4 Profiles of oxygen partial pressure in Column 3 during two series for times after dosing of 7.9, 24.5, 49.3, and 71.3 hours.

and bacterial-respiration rates) at a given time, but also on past conditions. Since quasi-steady state develops under the conditions of this experiment, the rate of oxygen addition by diffusion is equaled at each depth by the rate of deoxygenation from bacterial respiration.

Partial pressures of oxygen for samples taken about one hour after Column 1 was ponded with 500 ml of a 500-mg/l glucose solution and while the column was still ponded are presented in Table 5-1. The partial pressures, all less than 1.3 percent of those in the atmosphere (except for that at 60 cm), show that the column was essentially anaerobic during ponding. Some of the error can be attributed to contamination of the sample during withdrawal and analysis. One hour was about the minimum duration of ponding for Columns 1 and 3 after they had been dosed intermittently for about two months. These measurements therefore indicate that the atmosphere in the columns was devoid of oxygen just as infiltration was terminating.

Another series of gas samples was taken from Column 3 to study the distribution of oxygen before the quasi-steady state had developed. The data (Fig. 5-5) indicate that this condition had not been attained at about 3.5 hours after substrate addition. This time corresponded to that when drainage (infiltration and percolation) had essentially ceased. Measurements at 12 and 23 hours after

TABLE 5-1

Partial Pressures of Oxygen in a Ponded Column One Hour After Substrate Was Ponded

Depth (cm)	Partial Pressure (atm)
10	*
15	*
20	.0015
30	.0019
40	.0015
50	.0025
60	.0041

* Water rather than air was withdrawn by the sampling procedure

substrate addition show that the system was at a quasi-steady state. These measurements together with those of Fig. 5-4 show that the quasi-steady state was attained between 3.5 and 7.9 hours after substrate addition or between 2.5 and 6.9 hours after termination of infiltration.

Oxygen partial pressures were substantially higher at 3.5 hours than later because drainage allowed oxygen to enter the system by convection in addition to diffusion. At this time, the system could not be characterized by a quasi-

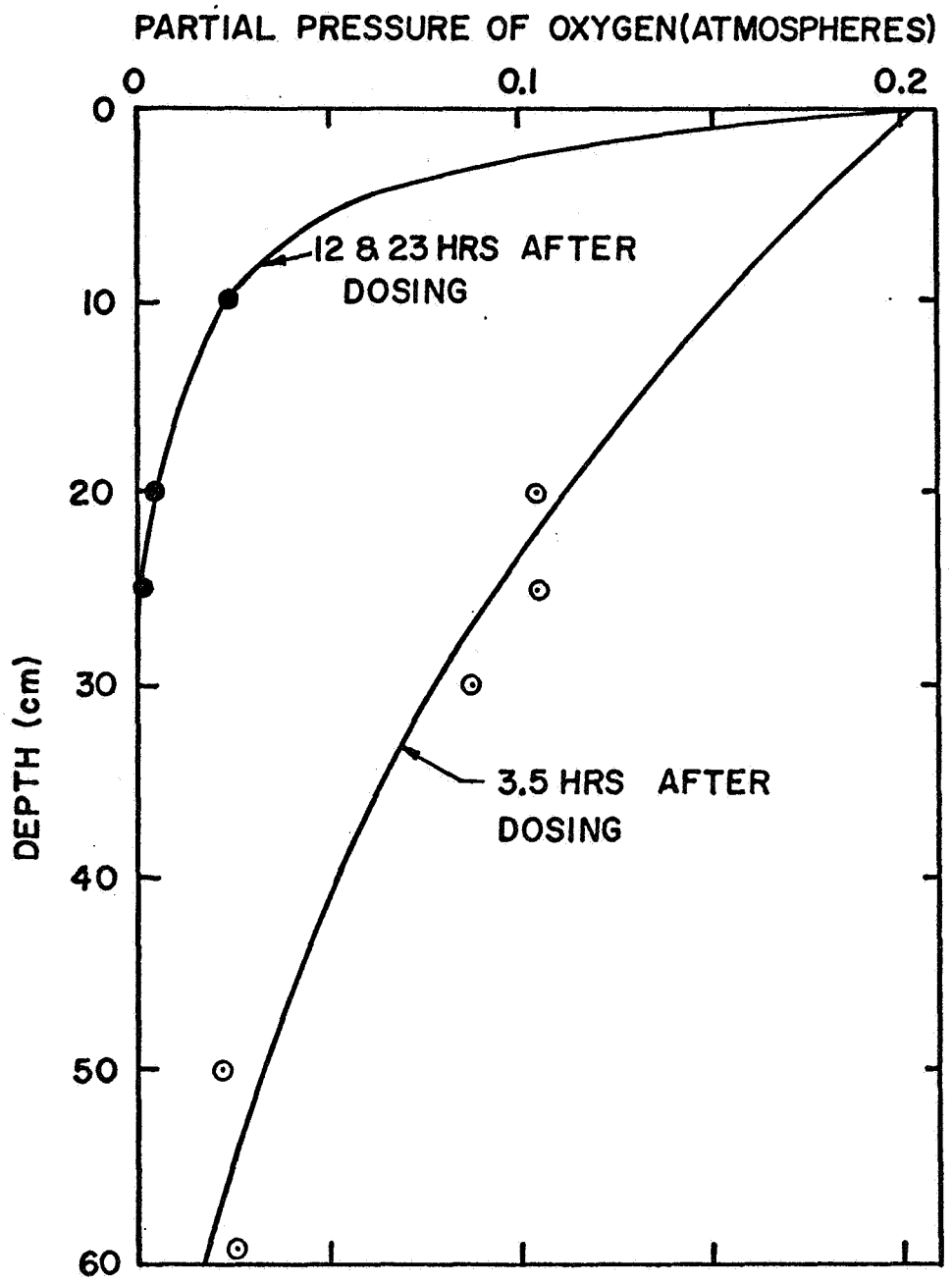


Fig. 5-5 Profiles of oxygen partial pressure in Column 3 for times after dosing of 3.5, 12, and 23 hours.

steady state because the oxygen profile depended on the past history rather than just on respiration rates and gas porosity at 3.5 hours. As drainage rates decreased, the oxygen partial pressures decreased to a quasi steady-state oxygen profile and then increased slowly with time.

5-2-2 Respiration Rates

Data calculated from measurements of respiration rates in Column 3 are shown in Fig. 5-6. The recorded data were first plotted for each Warburg flask on graphs such as Fig. 5-7 and slopes in terms of mm/hour obtained for times corresponding to those for the oxygen analyses (Fig. 5-2). Bacterial-respiration rates were then calculated by multiplying the slopes by the flask constant per gram of dry soil.

Most of the aerobic bacterial activity occurred near the surface, decreasing by more than an order of magnitude in about 40 cm and remaining roughly constant from 40 to 70 cm. Trends in Fig. 5-6 show that oxygen-uptake rates continuously decreased during the experiment.

It has been shown (44,45) that the rate of oxygen uptake for bacterial systems increases during the growth phase and that the beginning of decreased oxygen-uptake rate corresponds to the end of the growth phase. The decrease of oxygen uptake rates with time indicates that many microorganisms were already respiring more quickly

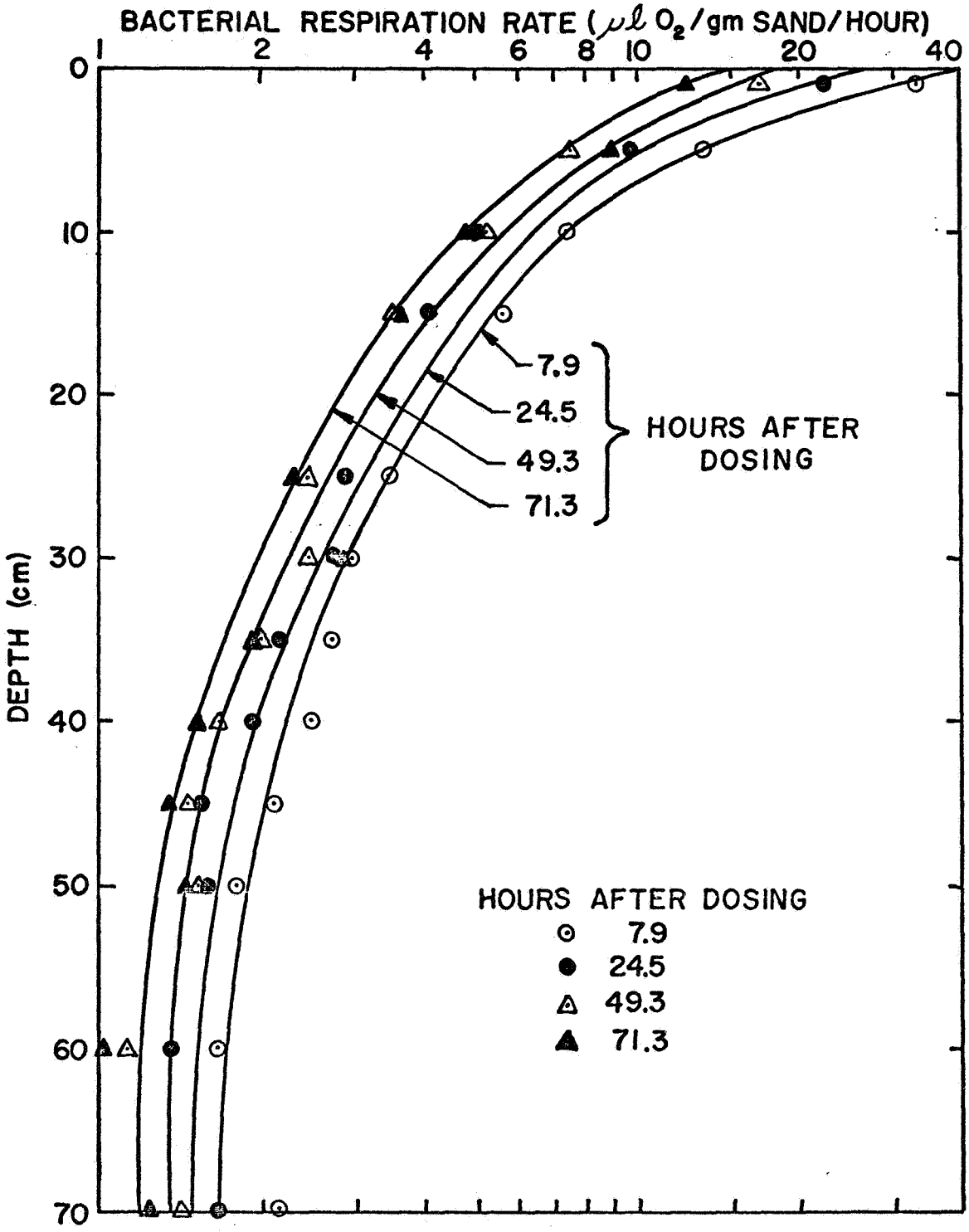


Fig. 5-6 Bacterial-respiration rates in Column 3 obtained with Warburg apparatus.

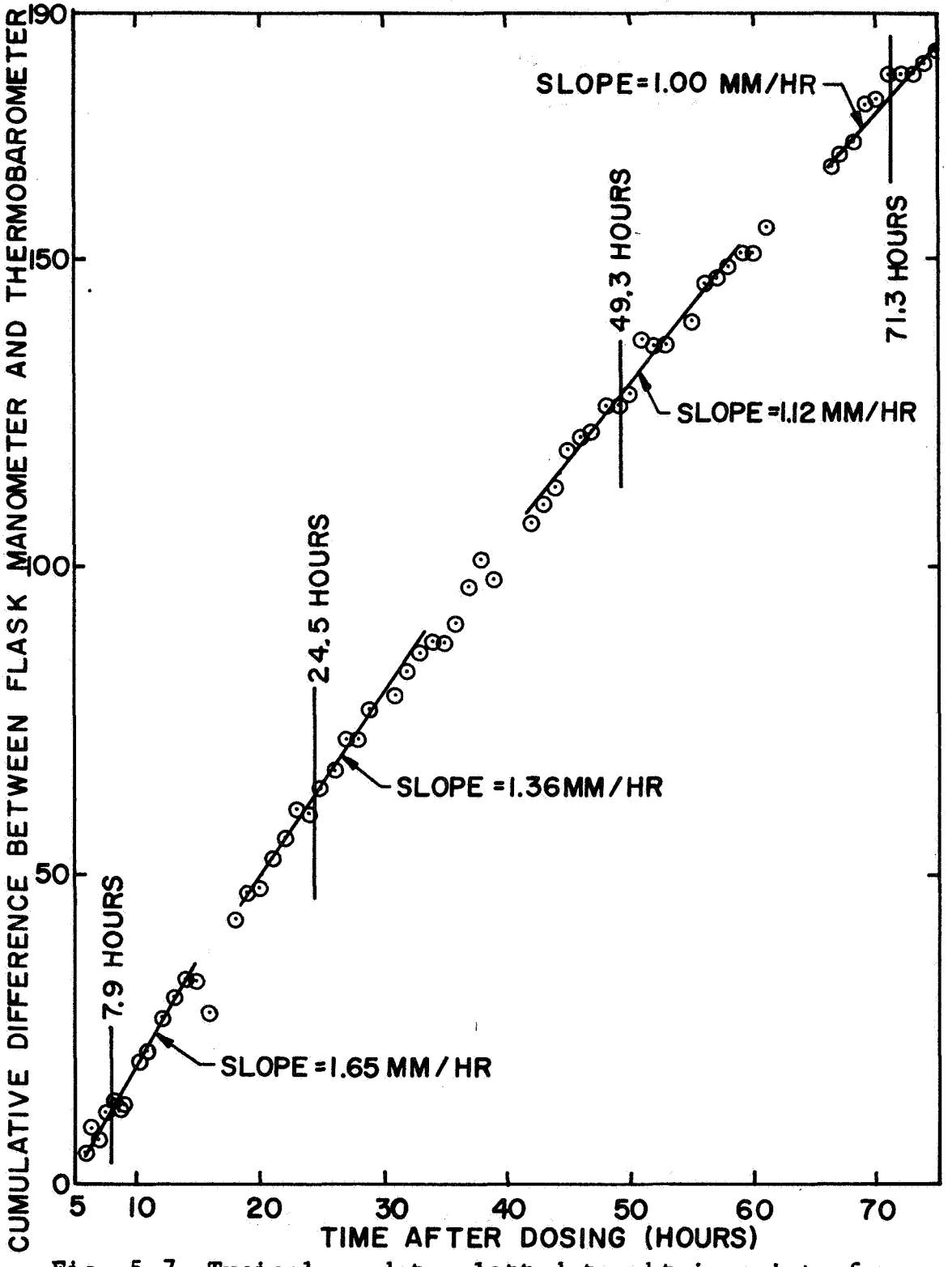


Fig. 5-7 Typical data plotted to obtain points for Fig. 5-6. Flask contained 25.04 grams of sand taken at depth of 60 cm in column. Flask constant was 11.35 μ l/mm.

than they were assimilating substrate when the Warburg measurements began about five hours after addition of substrate. Therefore, easily degradable substrate was no longer readily available to most bacteria.

5-2-3 Moisture Retention Measurements

A comparison of moisture profiles for 0.56 mm Ottawa sand when clean and after use in Column 3 is shown in Fig. 5-8. The data for clean sand were obtained with a fritted-glass Buchner funnel (Section 3-8-2). It should be noted that the ordinates of points plotted for the clean sand are distances above a water table and that the ordinate for the uppermost point is not a negative depth. The data for sand from Column 3 were obtained gravimetrically after disassembly. The latter points represent the condition about three hours after addition of substrate, when moisture movements had almost ceased.

Bacterial growth caused a large increase in moisture retention for the greater part of the column. The change in moisture retention near the surface is important because it diminishes the effective diffusivity for gases by decreasing the porosity.

5-2-4 Measurements of Redistribution of Moisture

Attenuation of a 1.5-inch diameter cylinder of water was determined from gamma-radiation measurements on an acrylic-plastic container with the same cross-section as

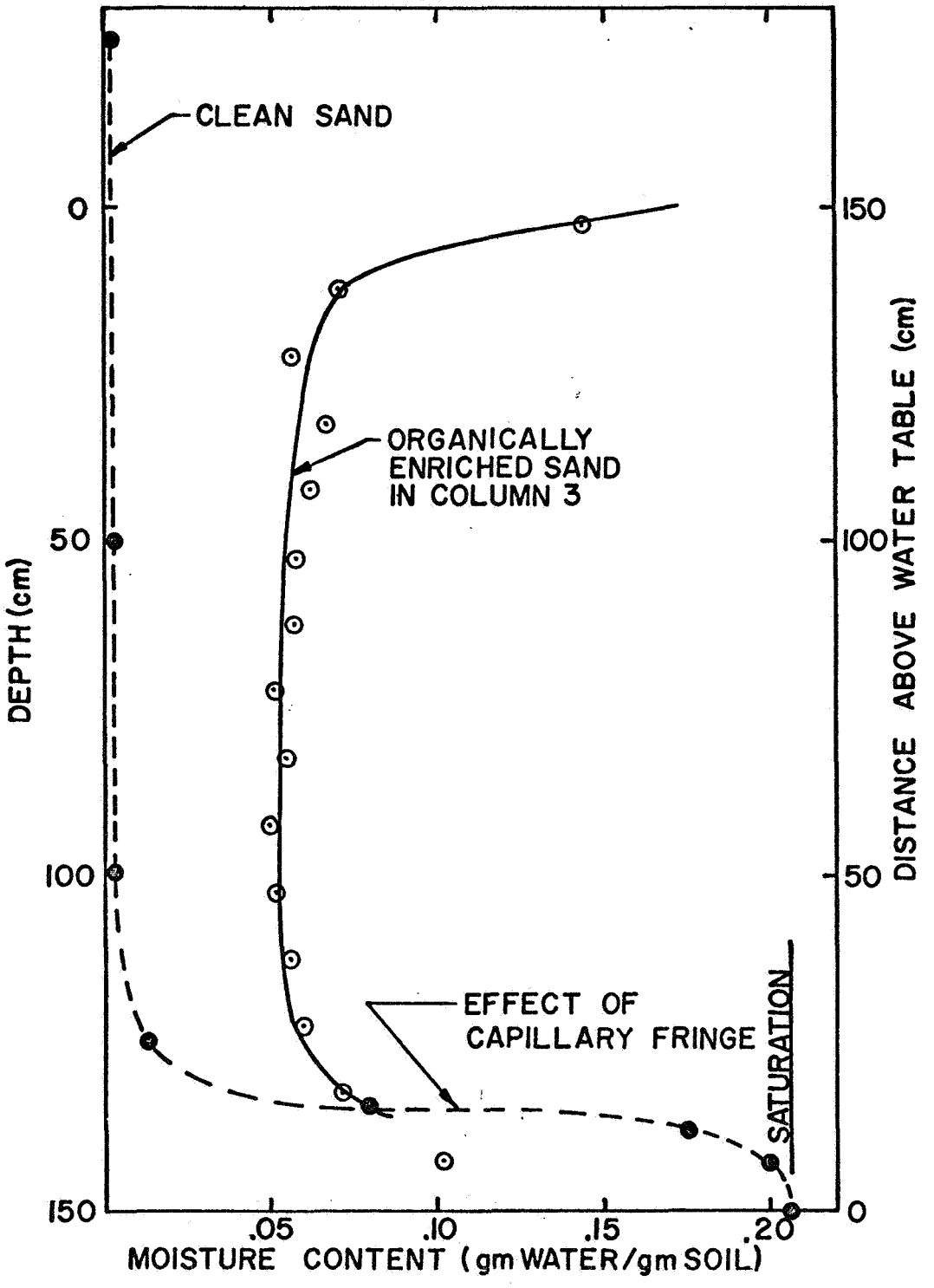


Fig. 5-8 Moisture profiles after drainage of clean Ottawa sand and Ottawa sand after use in Column 3.

the columns. The term $\mu_w z_w \rho_w$ in equation 3-3 for a 1.5-inch cylinder of water was found to be 0.314, corresponding to a mass adsorption coefficient (μ_w) of 0.0827 cm^2/gm for a water thickness of 1.5 inches. The value is within the range of 0.0815 to 0.0841 determined by others (33, 46).

Moisture changes during percolation of liquid were determined by measurements taken during four cycles. Different locations were selected each time. The results in Fig. 5-9 show no trend with depth. Although "noise" in the instrumentation caused some inaccuracy , the lack of an obvious drainage pattern can be attributed to the differences in the conditions near the surface, which conditions caused infiltration rates to vary considerably from day to day. The surface was scarified at intervals to maintain high infiltration rates. Even though the results are not consistent, some conclusions can still be drawn from the plots.

Information in Figs. 5-8 and 5-9 shows that water content before new infiltration was about 27 percent of saturation and increased to about 57 percent of saturation during infiltration for depths from 10 to 130 cm. Therefore, gas volume decreased from 73 to 43 percent of total pore volume in this distance, a volume decrease of about 160 ml. As a check, gas volume expelled at the column

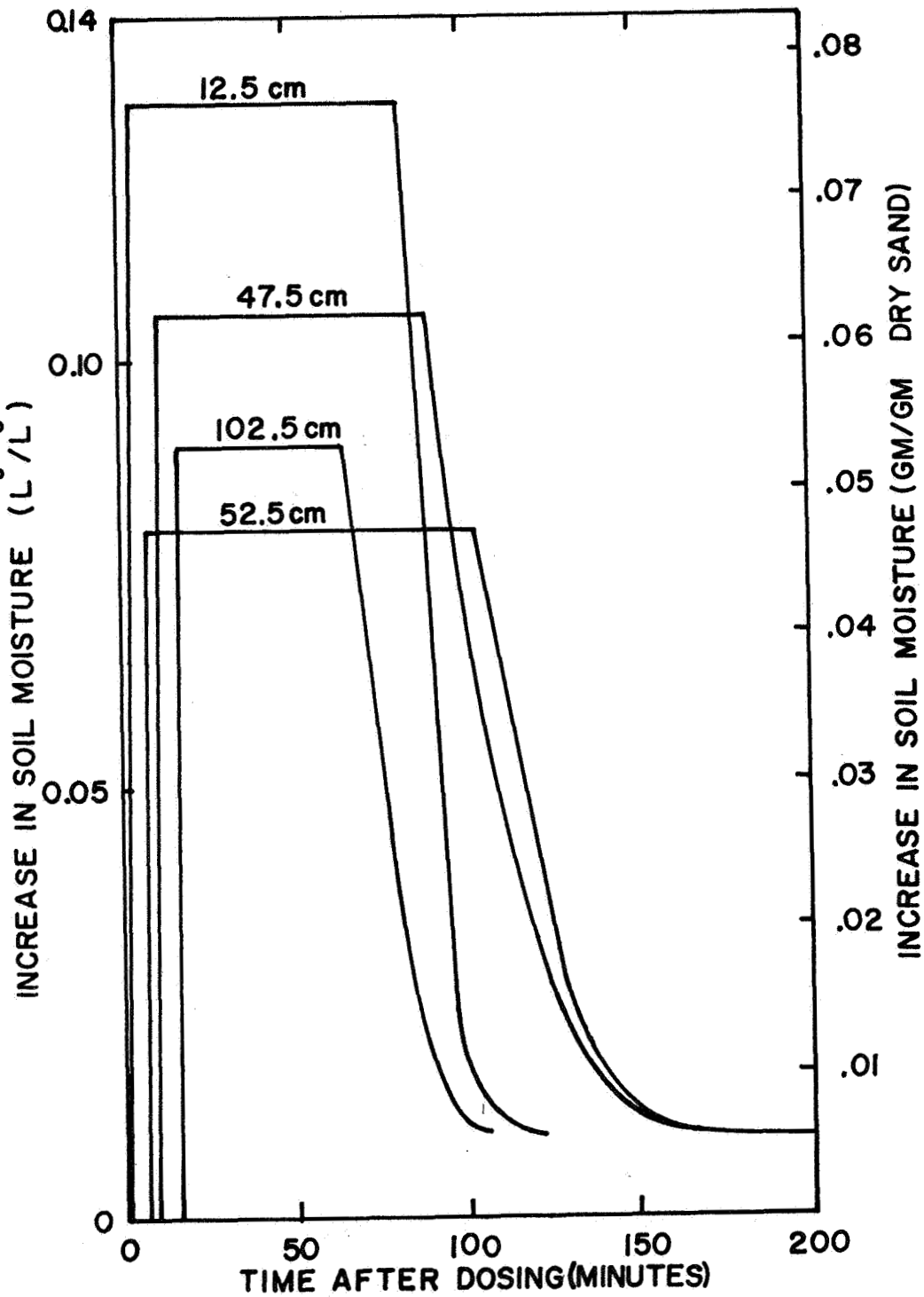


Fig. 5-9 Increase in soil moisture in Column 3 after addition of 44 cm of wastewater. Distances indicated near curves are depths from surface. Base line is moisture before dosing.

outlet was measured by water displacement and found to be about 140 ml. The total gas volume of the drained column can be calculated to be about 380 ml. Therefore, the water advancing in the column replaced only about 40 percent of the total gas volume.

For each case drawn in Fig. 5-9, the water content begins to decrease at the same time as infiltration is completed. The flow pattern was thus similar to that obtained by Biswas, et al (14) for redistribution in fine-textured soils, in which flow was saturated only at the surface and moisture contents decreased as ponding ceased (See Fig. 2-1b). The Ottawa sand used in these studies was not fine textured, but microorganisms and associated products in the column sufficiently reduced permeability at the surface so that the flow was unsaturated through most of the column. Similar behavior is to be expected in intermittent sand filters using other soils because growths should cause the minimum permeability to occur near the surface.

5-2-5 Analysis of Oxygen Transfer

The sets of curves in Fig. 5-4 for the two test series do not coincide because of differences in the surface condition of the column. The sand surface was scratched with a thin rod before substrate was added at the beginning of each series. The resulting condition of the sand matrix near the surface after the wastes had infiltrated was appar-

ently different in the two tests causing a change in total porosity, moisture retention, and therefore in effective diffusivity. Some of the difference in the oxygen profiles can be attributed to changes in respiration rates in the interval between the two sequences, but this difference is minor because respiration rate should change only slightly over a period of a few weeks after columns have been operating for several months.

In order to test the quasi steady-state model for oxygen transfer in intermittent sand filtration on the basis of measured parameters, equation 2-13 was used:

$$-\frac{d}{dz}\left(c\mathcal{D}\frac{dx_A}{dz}\right) = R_A \qquad (2\text{-}13)$$

During a series of measurements, the oxygen-concentration profile and the respiration rates changed, but the effective diffusivity at a given depth should have remained constant once moisture movements ended. The applicability of equation 2-13 to the problem can be determined, therefore, by using measured data on oxygen concentrations and respiration rates, and calculating the effective diffusivity. If the effective diffusivity at any given depth does not change with time, the model should be satisfactory.

To solve for effective diffusivity, equation 2-13 was integrated once and rearranged to yield

$$\mathcal{D}(z) = \frac{\int_z^L R_A(\xi)\, d\xi}{c\,\frac{dx_A}{dz}} \qquad (2\text{-}18)$$

Diffusivities during the two series in Fig. 5-4 were calculated by inserting into equation 2-18 respiration rates from Fig. 5-6 and slopes of the oxygen profiles in Fig. 5-4.

The distance L at which the partial pressure of oxygen becomes zero was first obtained from each of the eight curves in Fig. 5-4. The integral $\int_z^L R_A(\xi)d\xi$ was next calculated numerically with a digital-computer subroutine for integrating a plot function.

Slopes dx_A/dz of the curves fitted to the points in Fig. 5-4 were measured for the distances in the subroutine output, and the diffusivities calculated from equation 2-18. This process was repeated for each of the eight curves in Fig. 5-4 with the appropriate respiration rates from Fig. 5-6.

Calculated diffusivities for the four times in each of the two series are plotted in Fig. 5-10, with a curve drawn to fit the points. In both series, diffusivities are lower at the surface than elsewhere and increase with depth before becoming roughly constant. Diffusivities are low at the surface where bacteria reduce gas porosity because of a decrease of total pore volume by their bulk and their waste products and because of increased moisture retention. Fig. 5-10 indicates that bacterial growth can reduce the effective diffusivity at the surface to between 12 and 37 percent of that at about fifteen to thirty cm. The calculated diffusivities show that changing the sand structure near the

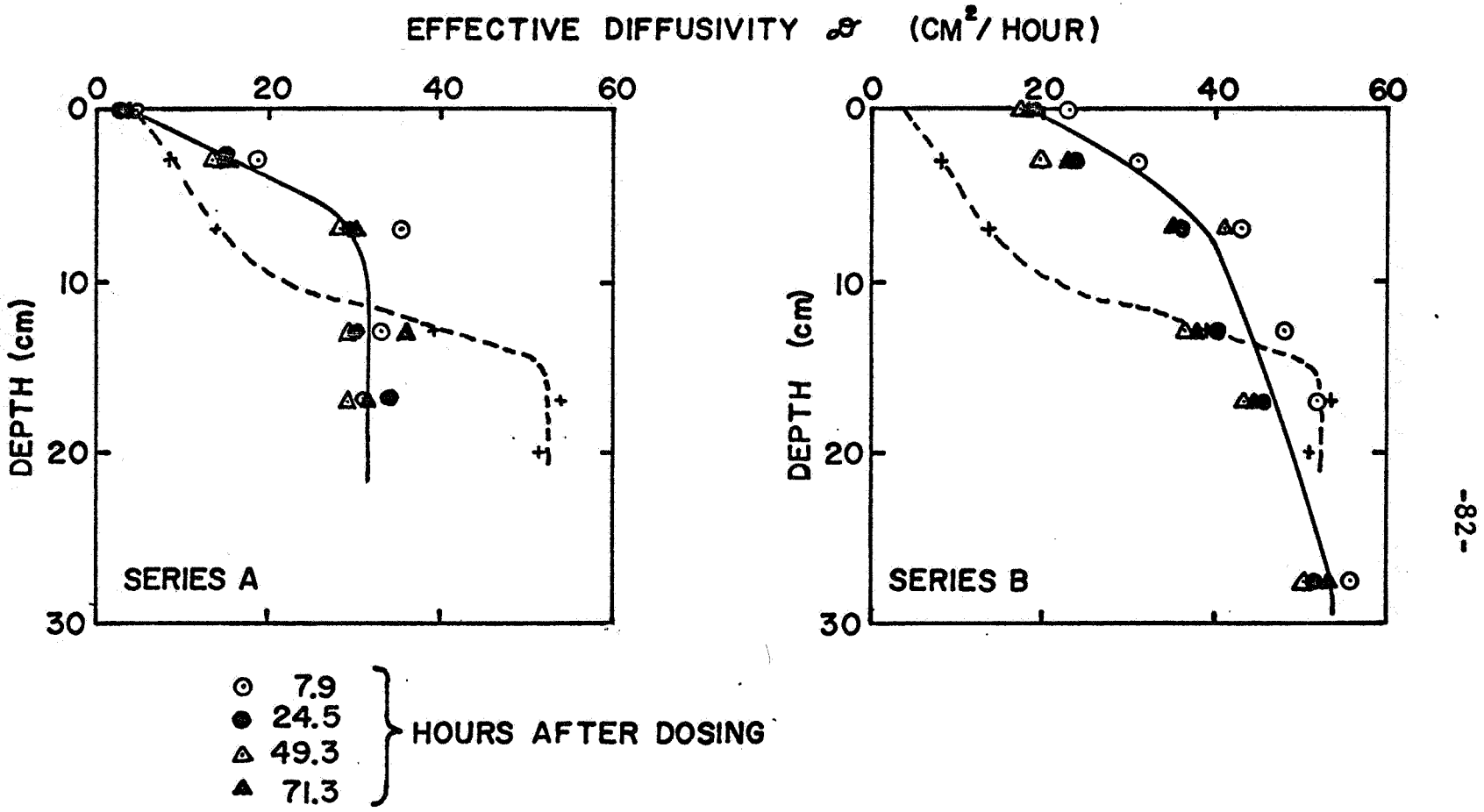


Fig. 5-10 Effective diffusivity calculated from quasi steady-state model (equation 2-13) using measured respiration rates (Fig. 5-6) and oxygen profiles (Fig. 5-4). Solid lines are fitted to data from model. Crosses fitted by dashed lines are calculated from Currie (27) data using moisture data from Fig. 5-8 and $\mathscr{D}_o$ = 806 cm^2/hr.

surface by scarification influenced the moisture retention (and, therefore, effective diffusivity) for a depth much greater than that of scarification. Effective diffusivities in Series A did not exceed about 30 cm^2/hour whereas in Series B they reached about 50 cm^2/hour.

A model is realistic only if the parameters calculated by it can be compared with at least some agreement to values calculated by an independent method. Diffusivities calculated using formulas taken from data plotted by Currie (27) can be used as the independent check (See Section 2-4). These diffusivities, with moisture contents from Fig. 5-8 and $\mathcal{D}_o$ = 806 cm^2/hour, are plotted as crosses fitted with dashed lines in Fig. 5-10. The results indicate that the diffusivities calculated by the model agree within a factor of two with those calculated on the basis of gas porosities.

The diffusivities calculated from respiration rates and oxygen profiles are all close to the fitted curve. Theoretically, there should be no scatter of the points at any depth during a series if moisture movements have stopped, but inaccuracies result from errors in measuring the slope of the oxygen profile and in determining oxygen uptake in the Warburg flasks. To determine if the scatter is caused by experimental error and not by error in the model, oxygen profiles were calculated using diffusivities from the curves in Fig. 5-10 and respiration rates from Fig. 5-6. The oxygen profiles were calculated using equation 2-19

$$x_A = x_{A0} + \frac{1}{c} \int_0^z \frac{\int_z^L R_A(\xi)\,d\xi}{\mathcal{D}(\xi)} \qquad (2\text{-}19)$$

which was solved numerically with the use of the IBM 7094 digital computer.

Because data for R_A were available only to 70 cm depth and it was felt that values of R_A might be needed for greater depths, estimates of R_A were added to the program for these depths. The estimates were 1.7, 1.5, 1.4, and 1.2 μl/gm/hr for times after dosing of 7.9, 24.5, 49.3, and 71.3 hours.

The same procedure was followed for each time to calculate variations of oxygen partial pressure with depth. Values of $\int_0^z R_A(\xi)\,d\xi$ for z ranging from 0 to 70 cm were calculated with a subroutine for integrating a plot function. Values of $\int_0^z R_A(\xi)\,d\xi$ for z greater than 70 cm were obtained by adding the area contributed by the estimated activity to that calculated at 70 cm.

The two boundary conditions for equation 2-13 were that $x_A = x_{A0}$ at the surface and that $dx_A/dz = 0$ at a depth L where aerobic bacterial respiration becomes zero because oxygen is absent. Because it was required that x_A also be zero at L this distance had to be estimated by trial and error. The scheme was to choose a trial of L of 10 cm and to calculate x_A from 0 to L. If x_A at L was positive, the next estimate of L was taken as 10 cm more and the process repeated until x_A at L was negative. A new L was then

chosen 9 cm less than the last value and x_A magnitudes calculated. A positive x_A at L caused a 1.0-cm increase in the trial L until x_A was negative at L. The depth L was reduced by 0.9 cm and then increased in 0.1-cm steps until x_A at L became negative. The first trial L for the next time for which experimental values were available was the last calculated L.

The value of $\int_0^L R_A(\xi)d\xi$ for the trial L was estimated by second-order interpolation. Diffusivities for the values of z used in the integration subroutine were estimated by interpolation.

The terms

$$\frac{\int_z^L R_A(\xi)d\xi}{\mathcal{D}(\xi)}$$

were calculated for the z's from the integration subroutine and

$$\int_0^z \frac{\int_z^L R_A(\xi)d\xi}{\mathcal{D}(\xi)}\, d\xi$$

obtained from the same subroutine. Oxygen partial pressures from 0 to L were then calculated from equation 2-19.

The results of the calculations (Fig. 5-11) except for 7.9 hours after dosing agree quite well with the oxygen concentrations measured in Column 3. They support the assumption of a quasi-steady state for the description of the oxygen profile and show that the change in respiration rates

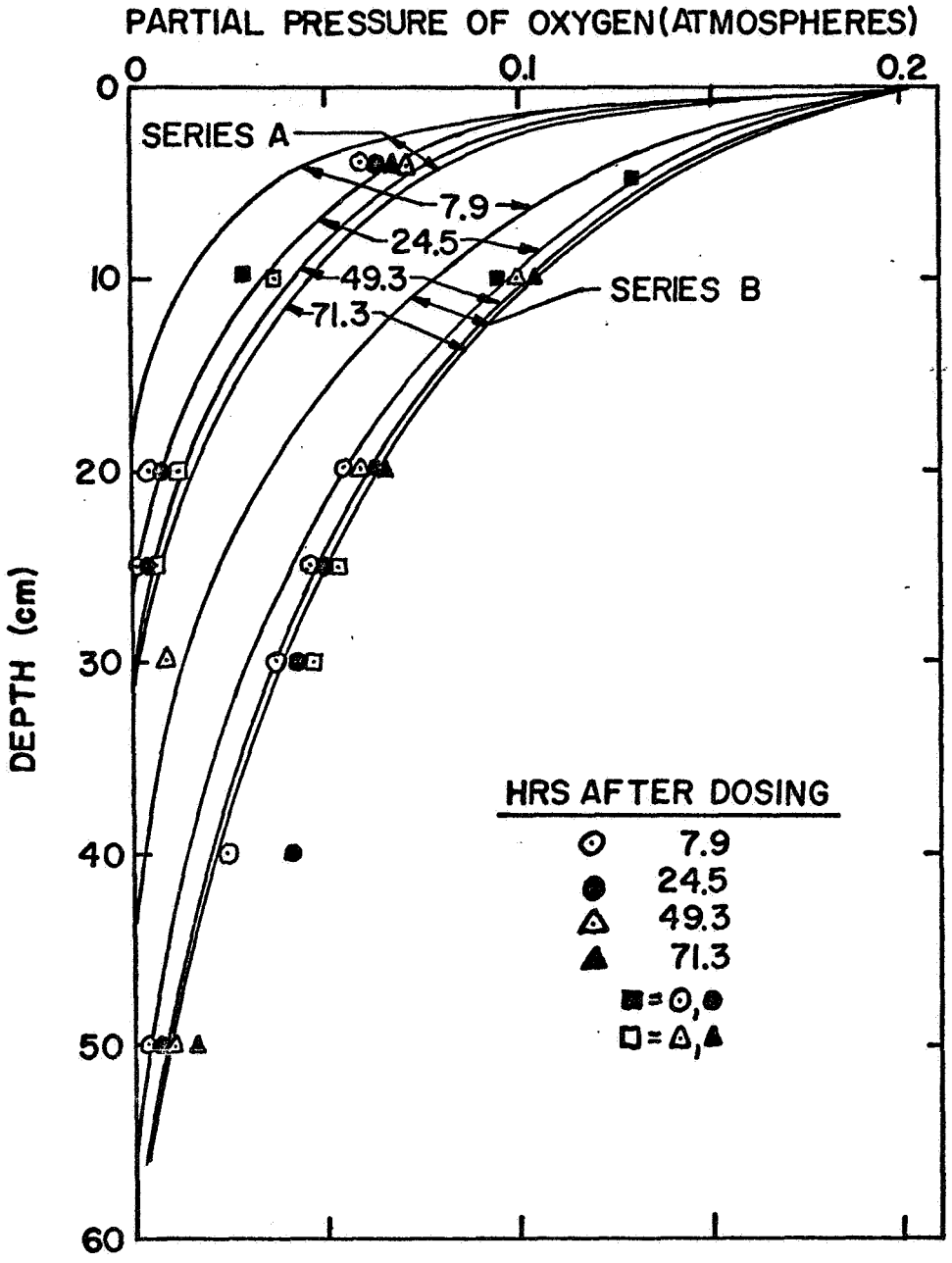


Fig. 5-11 Comparison of oxygen partial pressures calculated by quasi steady-state model with measured data.

is sufficient to alter the oxygen profile. The discrepancy at 7.9 hours after dosing can be attributed, at least in part, to the different test conditions. In the laboratory columns, oxygen does not reach some bacteria, especially near the point where two sand particles touch, because these bacteria are covered by other bacteria. Sanders (47) estimated that bacteria covered by more than two layers of other bacteria are limited by a lack of oxygen. The calculated curves are based on respiration rates obtained with a Warburg apparatus, in which sand particles tend to be separated. Hence, some bacteria that were covered by other layers in the sand columns became exposed to atmospheric oxygen when sand particles were separated and exerted a high initial oxygen-uptake rate.

No estimates of $\mathcal{D}$ and R_A are presently available for intermittent sand filters. Both values depend on the bacterial growth attained in the system, which growth in turn depends on many parameters, including composition and strength of wastewater, depth and frequency of ponding, and soil type. Since all these factors can be selected in designing and operating intermittent sand filters, their effect on $\mathcal{D}$ and R_A deserves more study.

5-2-6 Effluent Analyses

Effluent analyses on Columns 1 and 3 which were dosed with solutions of glucose, ammonium chloride, and salts

varied considerably during the study. Both columns initially formed nitrates, but other forms of bacteria apparently overgrew the nitrifying bacteria during the study because nitrate was not present in the effluent after four months of operation. It is not implied that glucose by its presence inhibited nitrification, although some organic compounds do inhibit. Glucose in concentrations less than 0.3M does not poison nitrifiers (48) but heterotrophic bacteria using it as substrate can assimilate all the ammonia present if initial ammonia concentration is low, so that no ammonia is available for nitrifying bacteria (49). Because ammonium was always present in the column effluent in this study, ammonium was available to nitrifying bacteria. The generation times for heterotrophic organisms are so much less than those of the nitrifiers (30 minutes vs 31 hours) that heterotrophic bacteria covered the nitrifying bacteria and prevented oxygen from reaching them.

Effluents from Columns 1 and 3 were collected during several cycles to determine the glucose present. Glucose removal was complete for the entire effluent collected during most cycles, but glucose was present in a few effluents. The latter results are of interest because they show the variation in removal with throughput when removal is not complete. Fig. 5-12 shows trends obtained

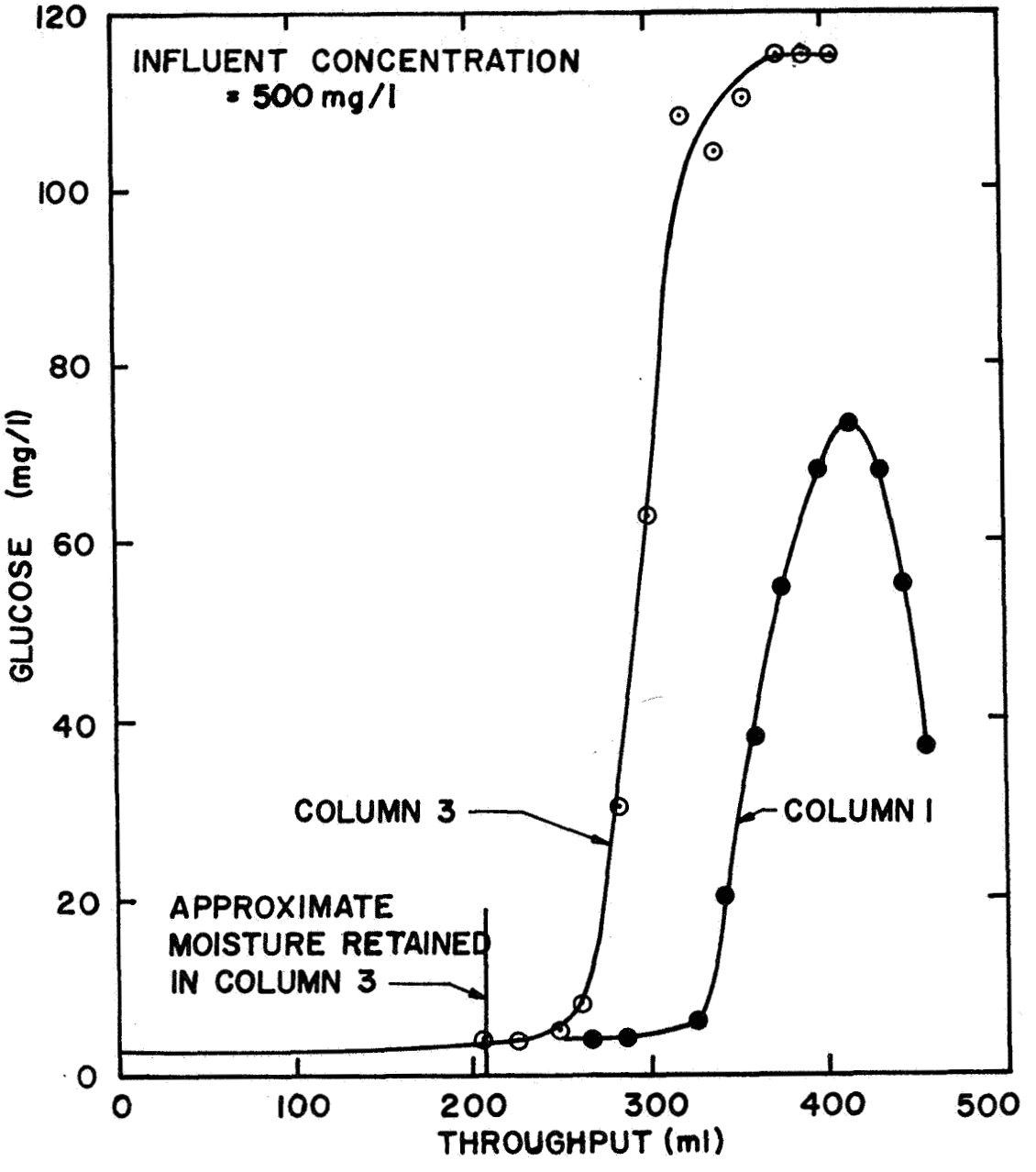


Fig. 5-12 Variation of glucose concentration with throughput from Column 3.

during cycles for which glucose was measured in the effluent. The effluent glucose was low at the beginning of drainage when the liquid that had been in the column for most of a cycle was being displaced. Glucose concentration then increased in the effluent for liquid that passed through the column while the column was anaerobic. The decrease for Column 1 after 420 ml indicates the effect of a reduction of flow rate, thus allowing more time for bacteria to metabolize glucose.

Considerable amounts of glucose were removed under anaerobic conditions because the effluent glucose concentrations in Fig. 5-12 are always much less than the influent concentration. It is generally assumed that adsorption of glucose on organic matter from bacterial growth is negligible compared to metabolism as a mechanism for rapid substrate removal in aerobic systems. This assumption is based on studies of activated sludge, such as those by Gaudy and Engelbrecht (44) and by Krishnan and Gaudy (50). Jeris and Cardenas (51) studied aerobic and anaerobic biodegradation of glucose in laboratory-scale units to which a daily dose of glucose was added. Results showed a zero-order reaction for both systems. The results for the aerobic system when extrapolated back to zero time agreed with the amount of glucose added within about three percent. Extrapolation for two

anaerobic systems resulted in values at time zero that were 7 to 24 percent less than the glucose added. The disagreement in the latter cases may have been caused by adsorption of glucose. Thus, it appears from the work by Jeris and Cardenas (51) that both adsorption and biodegradation can be responsible for rapid removal of glucose in anaerobic systems. Applied to this study, their work suggests that glucose was removed from the percolating liquid by adsorption and degradation.

Because nitrogenous material is an important fraction of the components of wastewaters and because no nitrate was formed in Columns 1 and 3, a column (Column 4) was refilled with 0.56 mm Ottawa sand and dosed with settled sewage to study nitrification. Gas analyses indicated that Column 4 remained aerobic down to the top of the capillary fringe. At Whittier Narrows, nitrification is prevented at some depths because the soil atmosphere lacks oxygen; in Column 4, nitrification occurred down to the top of the capillary fringe and to a certain extent within the fringe, where lack of oxygen also diminished nitrification.

Effluent from Column 4 was analyzed for Kjeldahl, nitrate, and nitrite nitrogen and for COD to determine their variation with throughput. The test for Kjeldahl nitrogen measures the concentration of nitrogen having

minus-three valence. The bacteria in the system served to oxidize the nitrogen to nitrite and nitrate, thereby lessening the Kjeldahl nitrogen present. Tests for the specific carbonaceous components in wastewaters would be preferred to the COD determination which is to measure the portion of organic matter that can be oxidized by a strong oxidant. Nevertheless, because of the large number of carbonaceous components in wastewaters, the COD test was used as an expedient.

The nitrogen measurements in Fig. 5-13 show large variation in effluent quality with throughput. The first increments collected, low in nitrate and high in Kjeldahl nitrogen, came from the capillary fringe where lack of oxygen limited nitrification. The nitrate peak and accompanying Kjeldahl-nitrogen valley correspond to liquid that had been in the aerobic portion of the column for almost a day. The position of the nitrate peak ahead of the total moisture retained in the column and the low nitrate concentration for larger throughput indicate that little nitrification occurred while liquid was draining. The decrease in Kjeldahl nitrogen during drainage was caused by filtration and adsorption of suspended and dissolved nitrogenous matter. Dispersion accounts for the nitrate nitrogen after drainage of a volume equal to the moisture in the column. Note that the ordinate values for

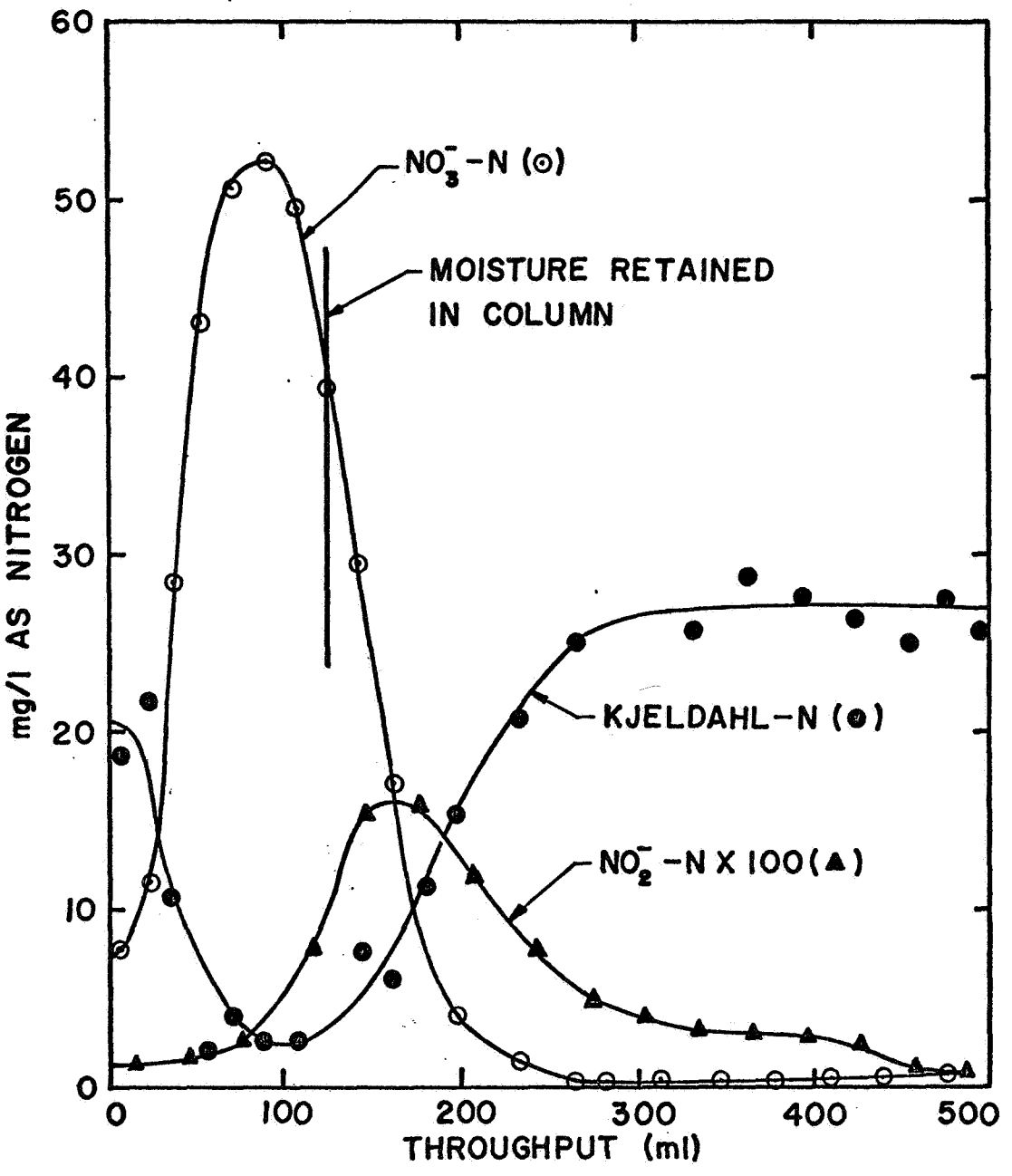


Fig. 5-13 Variation of nitrite, nitrate, and Kjeldahl nitrogen with throughput from Column 4.

nitrite have been multiplied by 100. The actual values are so small, all less than 0.15 mg/1, that they can be neglected in arriving at a nitrogen balance. No obvious reason has been found to explain the position of the nitrite peak after the nitrate peak, rather than at the same value of throughput.

The low COD for initial throughput (Fig. 5-14) results from displacement of liquid that was in the column for most of the cycle. COD content then rises with throughput because COD removal is limited during drainage. COD removal (Fig. 5-14) during column drainage was probably due mostly to filtration and adsorption rather than biodegradation because infiltration took only ten minutes. The settled sewage, even after passing through glass wool, still contained colloidal and particulate matter, which could be removed by the sand media. The decrease in COD after 380-ml throughput illustrates the improvement in COD removal when the percolate drains slowly and allows the wastes to remain in the column for a longer time.

Results from Column 4 in Figs. 5-13 and 5-14 can be compared with what would be expected from a spreading basin used for ground-water recharge. In a spreading basin, the wastewater passes through an aerobic portion a few feet deep and then trickles through an anaerobic zone

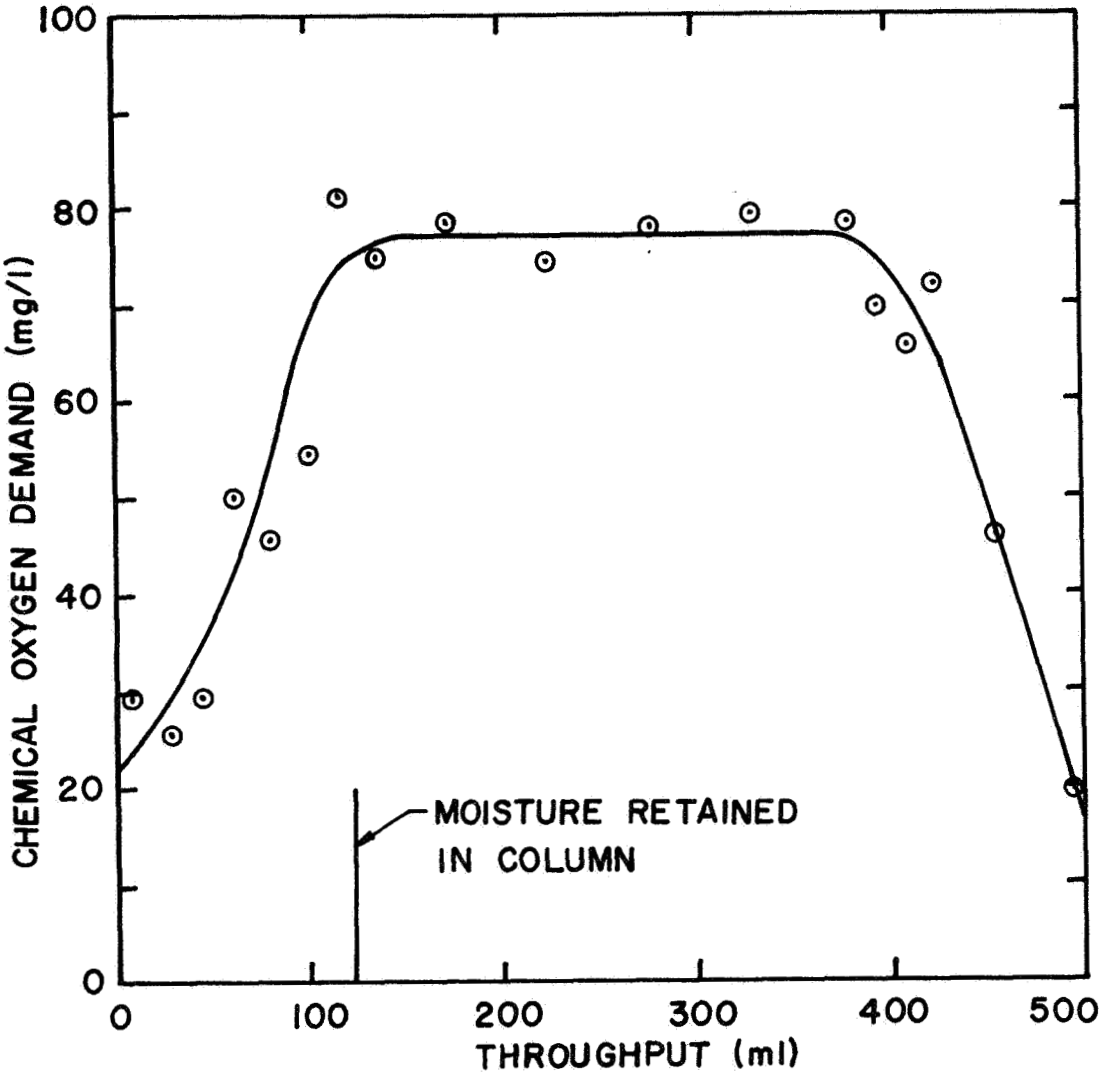


Fig. 5-14 Variation of chemical oxygen demand with throughput from Column 4.

to the water table. In the anaerobic zone, no further nitrification takes place, but some carbonaceous material may be removed from the percolate. Column 4 was similar except that the water table was brought up to the aerobic zone, and the percolate was collected just below the water table. Once collected, the percolate was not stabilized further, if it was immediately refrigerated. The similarity indicates that the variation of nitrate with throughput for a spreading basin should be comparable to the variation presented in Fig. 5-13 for a laboratory column. The COD reaching the water table below a spreading basin should be less than in the laboratory because bacterial activity in the anaerobic zone will diminish the carbonaceous matter.

5-2-7 Chemical Analyses of Sand Samples

Sand from Column 3 was analyzed for COD and for carbohydrate (glucose equivalent). The results (Fig. 5-15) along with those on bacterial respiration (Fig. 5-6) can be taken as indices of biological growth. They indicate that most of the bacterial growth occurs near the surface. This fact has been observed by many investigators using wastewaters, which contain colloidal and particulate matter as well as dissolved material. The measurements on Column 3 show that nutrient consisting of dissolved substrates only will also promote the same growth pattern.

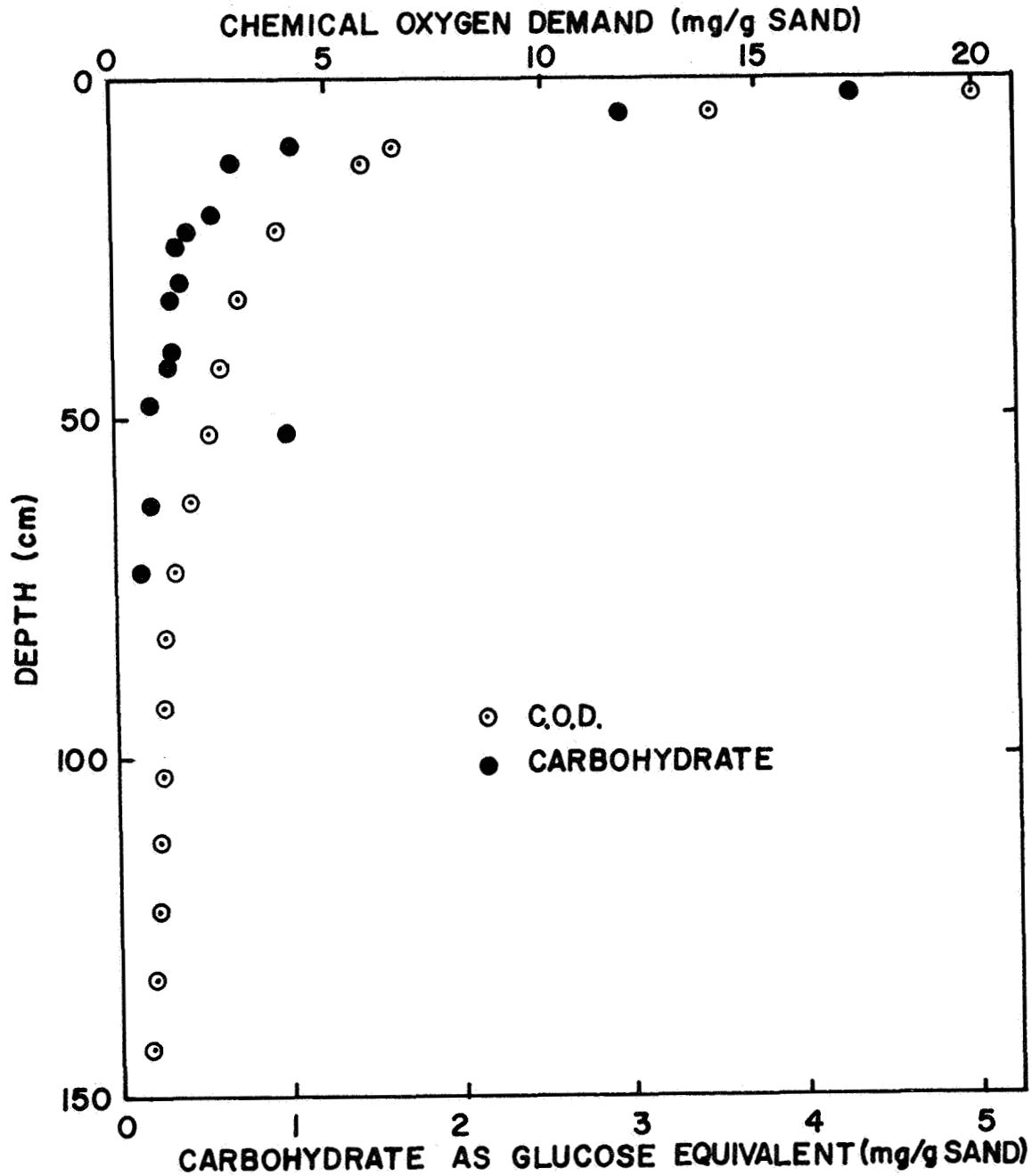


Fig. 5-15 Profiles of chemical oxygen demand and carbohydrate content of sand in Column 3.

Among the conditions affecting the distribution of bacterial growth in intermittent sand filtration, reduced oxygen concentration with increasing depth is most important, although other factors modify the growth pattern. Denser growth in the upper section of the sand column, whether caused at first by oxygen limitations at greater depth or by complete substrate removal in the upper section will decrease the permeability near the surface. Takagi (52) has shown that ponding water on a material of sufficiently low permeability above one of higher permeability will produce uniformly unsaturated flow in the lower section with the transition occurring within the region of low permeability. Once ponding ceases, the moisture probably drains as in Fig. 2-1b, with greater moisture content near the surface than elsewhere. This pattern allows substrate to be available for a longer time to organisms near the top, if all the easily oxidized substrate is stabilized during a cycle. Operation continued over many cycles further reduces the permeability near the surface and produces moisture retention as in Fig. 5-8. Because substrate is available near the surface for a longer portion of a cycle, more bacteria will be able to grow there.

Results (Fig. 5-16) for nitrate and nitrite extracted from sand in Column 4 twenty-four hours after substrate addition show similar trends for both anions, with nitrate concentrations roughly one hundred times nitrite concentrations. On Fig. 5-17 the nitrate is replotted in terms of μ g nitrate nitrogen per gram of dry sand. The increases in total nitrate at 0 to 10 cm and 120 to 140 cm correlate with increases in moisture content (Fig. 5-17). A larger moisture content, accompanied by larger total Kjeldahl nitrogen, allows substrate to be available to nitrifiers for a longer time during the cycle. Nitrifying organisms between 10 and 120 cm probably used up all of the readily available substrate. The decrease in total nitrate from 140 to 150 cm is attributable to the lack of oxygen. The increase between 0 and 10 cm above that from increased substrate volume was caused by availability of colloidal and particulate matter, which was removed by filtration.

The nitrate analyses on the soil extract indicate that 6900 μ g of nitrate nitrogen were produced during a cycle. However, the effluent analyses (Fig. 5-13) suggest that only 6140 μ g were produced. Denitrification does not seem to be the cause of disagreement because effluent analyses for nitrate and Kjeldahl nitrogen show a nitrogen balance (within two percent). Instead, a systematic error in one of the analyses may be the cause.

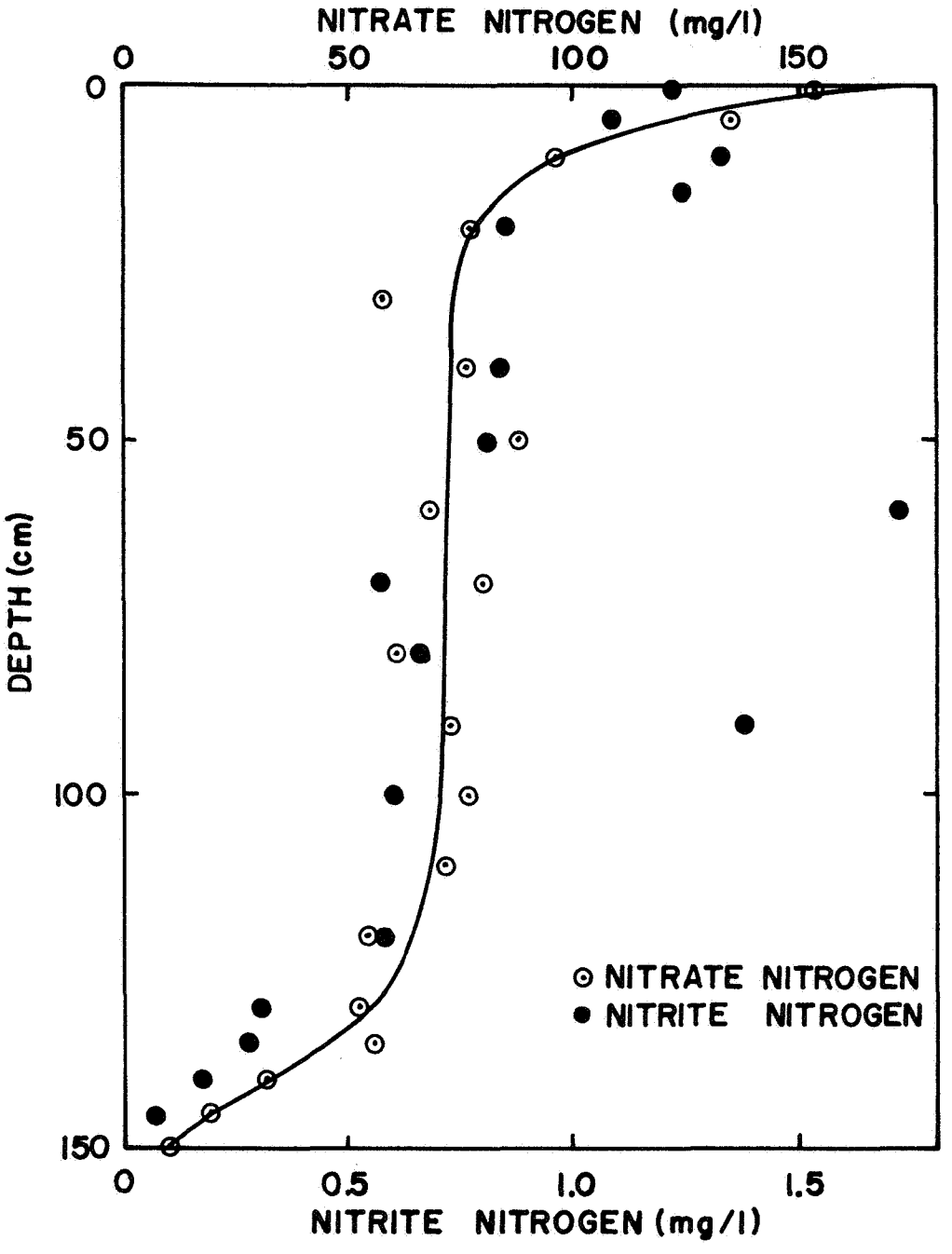


Fig. 5-16 Profiles of nitrate and nitrite concentrations in soil moisture in Column 4.

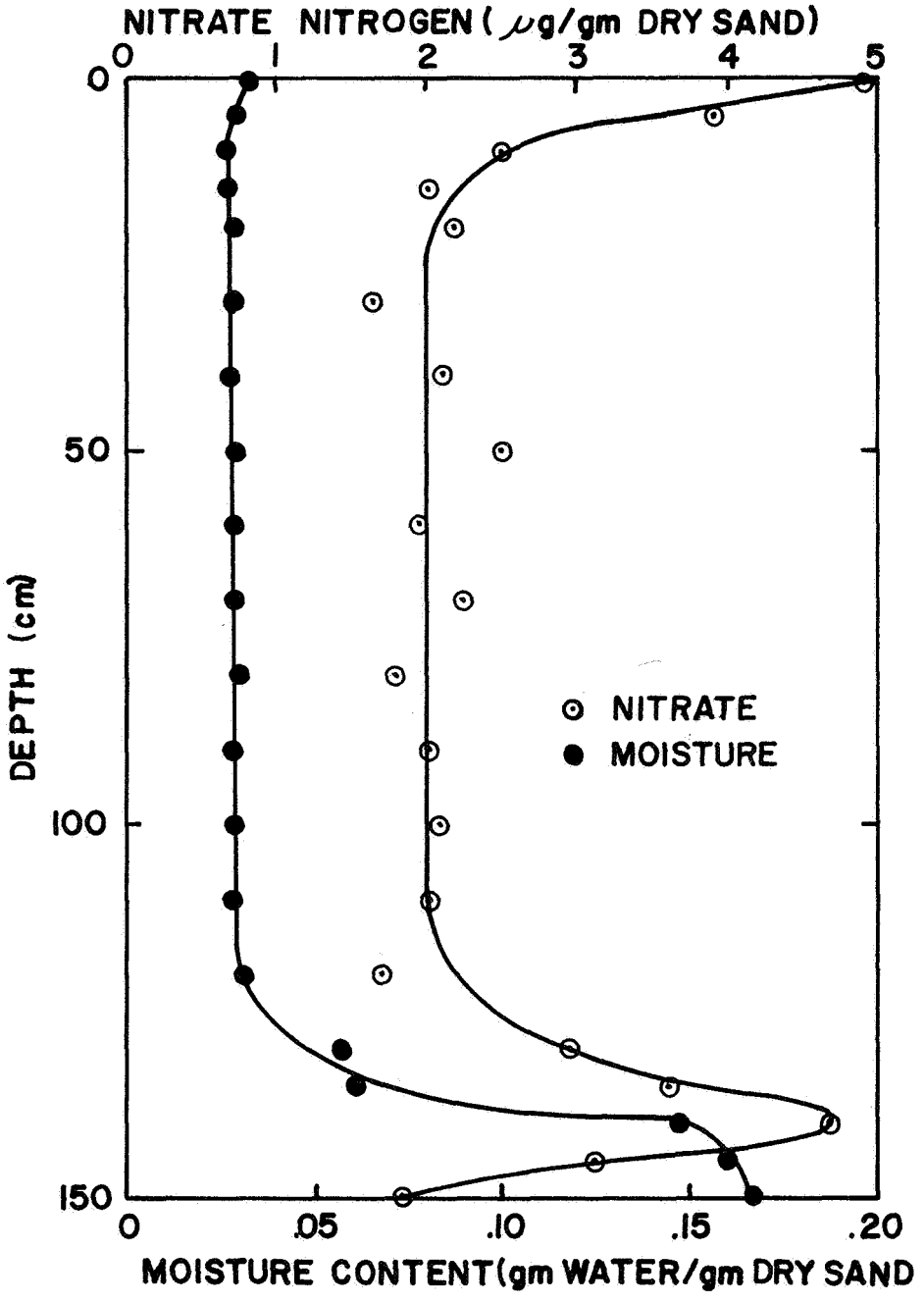


Fig. 5-17 Moisture profile after drainage of sand in Column 4 and profile of nitrate content per gram dry sand.

CHAPTER 6

SUMMARY AND CONCLUSIONS

6-1 Rationale of this Research

Although almost a hundred years of experience with intermittent sand filtration of wastewater had shown clearly that the system must be kept aerobic to prevent clogging, very little work had been done to study oxygen relationships in the system. For example, an extensive review of the literature did not uncover a single measurement of oxygen concentrations in the soil atmosphere for media used for treatment of sewage by intermittent percolation. The increasing use of spreading basins for tertiary treatment and ground-water recharge makes research on the oxygen relationships in soil systems pertinent inasmuch as oxygen availability may limit the usefulness of the method.

A simplified description of the various processes that occur during intermittent sand filtration was also needed because an understanding of the soil system as a whole is necessary to comprehend transfer and bacterial utilization of oxygen. A simplified description of the processes is useful by itself to explain variations in the quality of effluent from an intermittent sand filter.

6-2 Summary of Processes

6-2-1 Spreading Basin Operation

In practice, effluent from a primary or secondary wastewater treatment plant is spread on a basin of sand or soil and the area is allowed to drain until no liquid is ponded on the surface. The basin is then "rested" before further application of treated wastes to allow oxygen from the atmosphere to diffuse into the soil. Total cycle times (ponded plus resting) vary from about 4 hours to three weeks or more, with resting times constituting half or more of the total cycle.

6-2-2 Description of Processes

When effluent from a secondary treatment plant is applied to the surface of a spreading basin, pollutants are removed from the incoming liquid by adsorption on soil surfaces, by diffusion into stagnant zones, and by biological assimilation and synthesis. Concurrently, stabilized substances are returned to the percolate and move with it.

Biological activity within the sand or soil is limited while the surface of a spreading basin is ponded. Lack of oxygen limits the activity of aerobic organisms such as fungi and nitrifying bacteria. Anaerobic bacteria can degrade only portions of the available substrate because the liquid percolates through the soil too rapidly. Some components, such as alkyl benzene sulfonates, may not be

entirely removed from the percolate because the adsorbing surfaces become saturated.

After infiltration has ceased, oxygen reenters into the soil and aerobic bacterial activity progresses toward greater depths. The depth of aerobic bacterial activity is limited, however, because oxygen is depleted by biochemical utilization as air diffuses downward. Hence, some depths are always anaerobic. Consequently, not all percolate passes through the system with the same degree of treatment. Parts of the effluent that go beyond the maximum depth where oxygen is ever present receive no further aerobic treatment. It is possible, therefore, for percolate to show large variations with respect to the concentration of some contaminants, and some effluent portions show little change in these contaminants in the influent.

Nitrate can be used to indicate the extent of aerobic activity in the soil and to demonstrate the effects of displacement of pellicular water when wastewater is applied to the surface of a spreading basin. Because nitrification rates are low while the surface is ponded, the nitrate concentration in the soil water is roughly constant for some depth into the ground just as infiltration is completed. Nitrate is first formed near the surface and is formed at greater depths as oxygen becomes available. Nitrate cannot be produced, however, at depths where oxygen is absent.

It serves therefore to indicate the maximum depth to which oxygen penetrates.

Displacement of the nitrate formed during one cycle by newly applied wastewater is hypothesized to cause the percolate to have a nitrate wave (Fig. 2-3).

6-2-3 Oxygen Transport

The complexity of redistribution of moisture during intermittent percolation and the effect of such moisture redistribution on the movement of gas make any attempt at mathematically describing the oxygen concentration while moisture is still draining most difficult, if not impossible. It was observed, however, that after infiltration is completed, there developes a quasi-steady state, one in which the oxygen concentration does not depend on the past history of the media. This condition exists in the soil for several days after infiltration has stopped.

The differential equation describing the quasi-steady state is

$$-\frac{d}{dz}\left(\frac{1}{1-x_A+qx_A}\, cD\, \frac{dx_A}{dz}\right) = R_A \qquad (2\text{-}15)$$

for which notation is presented in Chapter 2. Because respiratory quotients (q) for intermittent sand filtration are not known, but probably lie between zero and one, equation 2-15 was solved for a simple model first with q equal to zero and then to one. The two solutions were reasonably close. It was thus decided to use equation 2-15

with a respiratory quotient equal to one to analyze data.

6-3 Principal Results

1. Analyses were conducted on soil samples from the Whittier Narrows Test Basin to determine the nitrate concentration in the soil water. As predicted by the foregoing discussion, the data show that only slight nitrification occurs while the basin is ponded and that the nitrate content in the soil moisture is roughly constant for some depth immediately after infiltration stops. Nitrification begins near the surface after infiltration ends and progresses downward as oxygen enters into the soil. The nitrate profile just before wastewater is added again to the surface shows high nitrate concentration for the first few feet and then a sharp drop in nitrate concentration where oxygen failed to penetrate and hence where no increase in nitrification occurred after infiltration.

2. Oxygen concentrations in gas samples taken from the soil atmosphere at the basin showed no trend with time but indicated that oxygen penetrated only to about two feet or less for times from 5 to 46 hours after completion of infiltration. Measurements taken at the same time to determine nitrate in soil water agreed with the oxygen measurements by showing that nitrification also took place only within the top two feet of the basin.

3. Oxygen profiles measured in laboratory columns dosed intermittently with a synthetic mixture showed only slight changes over more than 63 hours and could be described at sufficiently long times after substrate addition by a quasi steady-state equation for oxygen transport. The laboratory oxygen profiles and respiration rates obtained with a Warburg respirometer were used to calculate diffusivities from equation 2-18:

$$\mathcal{D}(z) = \frac{\int_z^L R_A(\xi)\,d\xi}{c\,\frac{dx_A}{dz}} \qquad (2\text{-}18)$$

The diffusivities were almost independent of time and were used to calculate new oxygen profiles which agreed quite well with the experimental profiles. Results indicated that scarifying the surface of the media influenced effective diffusivity for a depth much greater than that of scarification.

4. Data from gravimetric moisture determinations and from moisture determinations by gamma-radiation attentuation indicated that flow was always unsaturated throughout most of the column. Microorganisms and associated products at or near the surface of the column limited infiltration rates, thereby causing the lower areas to be unsaturated.

5. Incremental effluent samples were collected from a column dosed with glucose, ammonium chloride, and salts. Analyses for glucose in the effluent showed that glucose

removal was almost complete for the initial throughput in a cycle. Glucose concentrations, although increasing during further drainage, never approached the influent concentrations. Since the column became anaerobic during drainage, glucose was probably removed from the percolate by adsorption and anaerobic metabolism, as well as aerobically when oxygen was available.

6. A laboratory column dosed with settled sewage remained aerobic down to the capillary fringe. The first percolate after the capillary fringe was the most nitrified because it had been exposed to bacterial action for the longest time. The quality of the effluent then decreased until drainage rates dropped off sufficiently to allow more contact time between the system and substrate. Only slight nitrification occurred while the column was draining, although Kjeldahl nitrogen was diminished, probably by adsorption or filtration of nitrogenous substances.

7. Measurement of bacterial respiration rates and organic matter in laboratory columns showed that bacteria were most numerous near the surface. Their concentrations decreased rapidly with depth. Growth appeared to be limited mainly by a lack of oxygen.

6-4 Practical Application

In designing an intermittent sand filter, an engineer seeks a high rate of infiltration and a high degree

of treatment. He tries to attain these goals by proper design, which is achieved by choosing media having optimum characteristics, by determining the correct depth for the filter, and by selecting the desirable frequency and depth of wastewater application. These parameters also depend on the composition of the wastewater applied.

The results of this research cannot be applied directly to design a new facility; but they can be used to improve the operation of an existing installation or to aid in interpreting data from pilot-plant studies.

In most installations, observations of infiltration rates are used to determine the desired frequency and depth of application. No consideration is generally given to the quality of the percolate, if it is used for groundwater recharge, because of the difficulty in obtaining samples. Even when samples at various depths are collected in sampling pans, the results are questionable because the water so collected is not representative of the percolate in the soil. Clearly, the quality of the percolate is a major concern and should be determined when a groundwater basin is being recharged.

The maximum penetration of oxygen from the atmosphere into a typical spreading basin is only a few feet and it is almost constant for several days after a basin drains. A measure of this distance is valuable because it indicates the zone where nitrification and other aerobic activity

occur. The distance can be measured by withdrawing gas samples from the soil atmosphere and analyzing them for oxygen. It can also be determined by taking a soil boring and analyzing the soil moisture for nitrate or by obtaining liquid samples with porous cups for nitrate analysis. Nitrate is high in the aerobic zone and decreases sharply where the soil becomes aerobic.

The maximum volume applied per cycle should be equal to the moisture retained in the aerobic zone after drainage to insure that all the wastewater will receive aerobic treatment. The frequency of application for complete nitrification can be determined by adjusting the frequency until analyses from core samples or from samples extracted through porous cups indicate that nitrification is essentially complete. Tests other than those for nitrate can be used whenever there is reason to believe that the concentration of some other contaminant is critical.

In pilot-plant studies and in installations where underdrains collect the percolate, gas determinations can be used to measure the oxygen profile. However, analyses of incremental samples of percolate also yield good information. Most studies on these systems have collected the entire percolate or a composite sample for chemical analysis. This procedure is poor because it yields little information suggesting how the system should be operated for better results. Incremental samples should be collected

and analyzed to observe the variation of percolate quality with throughput.

Because oxygen is available only for the first few feet, samples collected from greater depths will not be high in oxidized products, such as nitrate, just after the surface is ponded. Percolate at greater depths will have passed through the upper few feet too quickly to become oxidized or while ponding caused the distance to become anaerobic. Nitrate and other oxidized products in the percolate at greater depths will increase as moisture retained at the top of the filter is displaced by the incoming liquid. Oxidized products will then decrease as newly added wastewater percolates to the bottom of the column or to the underdrain. A plot of the variation of oxidized products, such as nitrate, with throughput over several cycles will reveal a series of peaks. The volume added each cycle should be reduced until the volumes between the nitrate peaks become very small. The frequency of application should be adjusted to the maximum that will completely nitrify the volume retained.

The optimum media to be used in intermittent sand filters cannot be determined on the basis of this research. Some criteria for this media do present themselves, however. The media must be fine enough that its surface will be large and therefore support abundant microbial growth. On

the other hand, it must be coarse enough to allow a high effective diffusivity for gases. Coarse media do not retain as much moisture as fine-textured media and maintain higher porosities when drained.

Depth to the water table or to underdrains should be enough that the capillary fringe is below the aerobic zone and does not decrease the effective porosity of that zone.

6-5 Suggestions for Further Studies

1. This investigation has used nitrification as a major index of the efficiency of intermittent sand filters. Other components in reclaimed wastewaters may be more important, especially if they may be harmful to people drinking reclaimed water. Further research might well be conducted to determine the behavior of many more compounds than have been measured to date in wastewaters. Their removal and stabilization by percolation through soil systems should be determined.

2. Research is needed to allow engineers to design intermittent sand filters properly without the need for pilot-plant studies. Effective design also includes decisions relative to frequency and depth of application before the installation is built. Studies are necessary to determine the effects of variations in characteristics of porous media and wastewater composition on respiration rates. These data are needed to apply equation 2-13 to the

design. Variation of substrate utilization with soil type also deserves consideration.

3. In one of the laboratory columns (No. 3), nitrification ceased when higher concentrations of glucose were added to the substrate. It is evident that heterotrophic bacteria outgrew the autotrophic nitrifying bacteria. When large quantities of carbonaceous substrate were added, it appears that carbonaceous bacteria were not forced to respire endogenously and consequently they multiplied heavily. Their generation times are so much less than those of the nitrifiers (30 minutes vs. 31 hours) that they completely enveloped the nitrifiers. These observations require further study.

NOTATION

Dimensions are given in terms of mass (M), length (L), and time (t). Symbols that appear infrequently or in one section only are not listed.

c = total molar concentration, mols/L^3

c_A = molar concentration of species A, mols/L^3

$\mathcal{D}$ = effective diffusivity, L^2/t

$\mathcal{D}_o$ = diffusivity in free space, L^2/t

I = measured γ-radiation with interference

I_0 = measured γ-radiation without interference

K = capillary conductivity, L/t

L = depth at which oxygen concentration becomes zero, L

N_{Az} = molar flux of gas A in z direction with respect to stationary coordinates, mols/L^2t

p = capillary head, L

q = respiratory quotient, dimensionless

R_A = molar rate of production of gas A, mols/tL^3

t = time, t

x_A = mole fraction of gas A, dimensionless

z = thickness of material, or soil depth, L

ϵ = gas porosity, dimensionless

θ = soil water content, L^3/L^3

μ = mass absorption coefficient of absorbing material for given energy of radiation, L^2/M

ρ = density of material, M/L^3

ρ' = bulk density of material, M/L^3

REFERENCES

1. Sedgwick, W.T. 1902. Principles of Sanitary Science and the Public Health. MacMillan, New York.

2. Kinnicutt, L.P., C.-E.A. Winslow, and R.W. Pratt. 1913. Sewage Disposal. Wiley, New York.

3. Amramy, A. 1965. Waste treatment for ground water recharge. Air & Wat. Pollut. Int. J. 9:605-620. Also, in Advances in Water Pollution Research, Proceedings of the Second International Conference. Pergamon, London. 2:147-160.

4. Levin, G.V. 1967. The new pollution. Civil Engineering. 37:(No. 5) 68-71.

5. Allison, L.E. 1947. Effect of microorganisms on permeability of soil under prolonged submergence. Soil Sci. 63:439-450.

6. Gupta, R.P. and D. Swartzendruber. 1962. Flow-associated reduction in the hydraulic conductivity of quartz sand. Soil Sci. Soc. Amer. Proc. 26:6-10.

7. McGauhey, P.H. and J.H. Winneberger. 1964. Studies of the failure of septic tank percolation systems. J. Water Pollution Control Federation. 36:593-606.

8. Avnimelech, Y. and Z. Nevo. 1964. Biological clogging of sands. Soil Sci. 98:222-226.

9. Mitchell, R. and Z. Nevo. 1964. Effect of bacterial polysaccharide accumulation on infiltration of water through sand. Applied Microbiol. 12:219-223.

10. Thomas, R.E., W.A. Schwartz, and T.W. Bendixen. 1966. Soil chemical changes and infiltration rate reduction under sewage spreading. Soil Sci. Soc. Amer. Proc. 30:641-646.

11. Robeck, G.G., T.W. Bendixen, W.A. Schwartz, and R.L. Woodward. 1964. Factors influencing the design and operation of soil systems for waste treatment. J.Water Pollution Control Federation. 36:971-983.

12. Laverty, F.B., R. Stone, and L.A. Meyerson. 1961. Reclaiming Hyperion effluent. J. Sanit. Eng. Division, Proc. Amer. Soc. Civil Eng. 87:(SA6)1-40.

13. McMichael, F.C. and J.E. McKee. 1966. Final Report of Research on Wastewater Reclamation at Whittier Narrows. California State Water Quality Control Board, Publication No. 33.

14. Biswas, T.D., D.R. Nielsen, and J.W. Biggar. 1966. Redistribution of soil water after infiltration. Water Resources Res. 2:513-524.

15. Wang, F.C. 1963. An approach to the solution of unsteady unsaturated flow problems in soils. Technical Report No. 19, Department of Civil Engineering, Stanford University.

16. Whisler, F.D. and A. Klute. 1965. The numerical analysis of infiltration, considering hysteresis, into a vertical soil column at equilibrium under gravity. Soil Sci. Soc. Amer. Proc. 29:489-494.

17. Fujioka, Y. and T. Kitamura. 1964. Approximate solution of a vertical drainage problem. J. Geophys. Res. 69:5249-5255.

18. Gardner, W.R. 1962. Approximate solution of a non-steady state drainage problem. Soil Sci. Soc. Amer. Proc. 26:129-132.

19. Youngs, E.G. 1960. The drainage of liquids from porous materials. J. Geophys. Res. 65:4025-4030.

20. Thimann, K.V. 1963. The Life of Bacteria, 2nd ed. MacMillan, New York.

21. Schön, G. 1965. Untersuchungen über den Nutzeffekt von Nitrobacter winogradskyi Buch. Arch. Mikrobiol. 50:111-132.

22. Broadbent, F.E. and F. Clark. 1965. Denitrification. Soil Nitrogen. American Society of Agronomy, Madison, Wisconsin. :344-359.

23. Camp, T.R. 1963. Water and Its Impurities. Reinhold, New York.

24. Morgan, G.B. and F.W. Gilcreas. 1960. Distribution of carbohydrate metabolism in sewage sand filters. Water and Sewage Works. 107:485-488.

25. Bird, B.B., W.E. Stewart, and E.N. Lightfoot. 1960. Transport Phenomena. Wiley, New York.

26. Currie, J.A. 1960. Gaseous diffusion in porous media. Part 2-Dry granular materials. Br. J. Appl. Phys. 11:318-324.

27. Currie, J.A. 1961. Gaseous diffusion in porous media. Part 3-Wet granular materials. Br. J. Appl. Phys. 12:275-281.

28. White, A., P. Handler, and E.L. Smith. 1964. Principles of Biochemistry, 3rd ed. McGraw-Hill, New York.

29. Papendick, R.I. and J.R. Runkles. 1965. Transient-state oxygen diffusion in soil: I The case when rate of oxygen consumption is constant. Soil Sci. 100:251-261.

30. Van Bavel, C.H.M. 1951. A soil aeration theory based on diffusion. Soil Sci. 72:33-46.

31. Ritchie, J.T. 1964. Soil aeration characterization using gas chromatography. Ph.D. Thesis, Iowa State University of Science and Technology.

32. Kimball, B.A. 1965. Soil atmosphere composition as related to microbial activity and diffusion coefficient. M.S. Thesis, Iowa State University of Science and Technology.

33. Davidson, J.M., J.W. Biggar, and D.R. Nielsen. 1963. Gamma-radiation attenuation for measuring bulk density and transient water flow in porous materials. J. Geophys. Res. 68:4777-4783.

34. Taylor, E.W. 1958. The Examination of Waters and Water Supplies (Thresh, Beadle & Suckling), 7th ed. Little Brown, Boston.

35. American Public Health Association. 1965. Standard Methods for the Examination of Water and Wastewater, 12th ed. New York.

36. Bremner, J.M. 1965. Inorganic forms of nitrogen. Methods of Soil Analysis, Part 2. American Society of Agronomy, Madison, Wisconsin: 1179-1237.

37. Gaudy, A.F., Jr. 1962. Colorimetric determination of protein and carbohydrate. Industrial Water and Wastes. 7:17-22.

38. Brink, R.H., Jr., P. Dubach, and D.L.Lynch. 1960. Measurement of carbohydrates in soil hydrolyzates with anthrone. Soil Sci. 89:157-166.

39. Umbreit, W.W., R.H. Burris, and J.F. Stauffer. 1964. Manometric Techniques, 4th ed. Burgess, Minneapolis.

40. Vomocil, J.A. 1965. Porosity. Methods of Soil Analysis, Part 1. American Society of Agronomy, Madison, Wisconsin.

41. McMichael, F.C. 1966. Unpublished studies at California Institute of Technology.

42. Krone, R., H.F. Ludwig, and J.F. Thomas. 1952. Porous tube device for sampling soil solutions during water spreading operations. Soil Sci. 73:211-219.

43. Miller, R.J., J.W. Biggar, and D.R. Nielsen. 1965. Chloride displacement in panoche clay loam in relation to water movement and distribution. Water Resources Res. 1:63-73.

44. Gaudy, A.F. Jr. and R.S. Engelbrecht. 1961. Quantitative and qualitative shock loading of activated sludge systems. J. Water Pollution Control Federation. 33:800-816.

45. McWhorter, T.R. and H. Heukelekian. 1964. Growth and endogenous phases in the oxidation of glucose. Proc. 1st Inter. Conf. on Water Poll. Control, MacMillan, New York, 2: 419-436

46. Watson, K.K. 1965. Non-continuous porous media. Report No. 84. University of New South Wales Water Research Laboratory.

47. Sanders, W.M., III. 1967. The growth and development of attached stream bacteria, part 1. Theoretical growth kinetics of attached stream bacteria. Water Resources Res. 3:81-87.

48. Meiklejohn, J. 1954. Some aspects of the physiology of the nitrifying bacteria. Autotrophic Micro-organisms. University Press, Cambridge: 68-83.

49. DeMarco, J., J. Kurbiel, J.M. Symons, and G. Robeck. 1967. Influence of environmental factors on the nitrogen cycle in water. Jour. Amer. Water Works Assn. 59:580-592.

50. Krishnan, P. and A.F. Gaudy, Jr. 1966. Mechanism and kinetics of substrate utilization at high biological solids concentrations. Proc. 21st Ind. Waste Conf., Purdue Univ.:495-510.

51. Jeris, J.S. and R.R. Cardenas, Jr. 1966. Glucose disappearance in biological treatment systems. Appl. Microbiol. 14:857-864.

52. Takagi, S. 1960. Analysis of the vertical downward flow of water through a two-layered soil. Soil Sci. 90:98-103.